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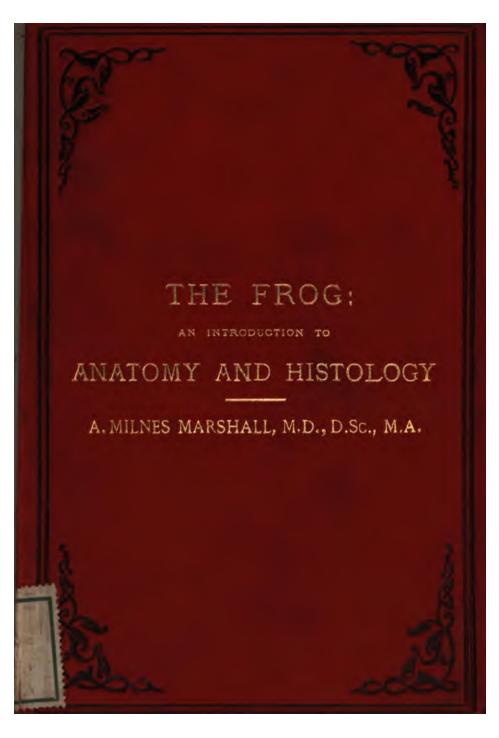
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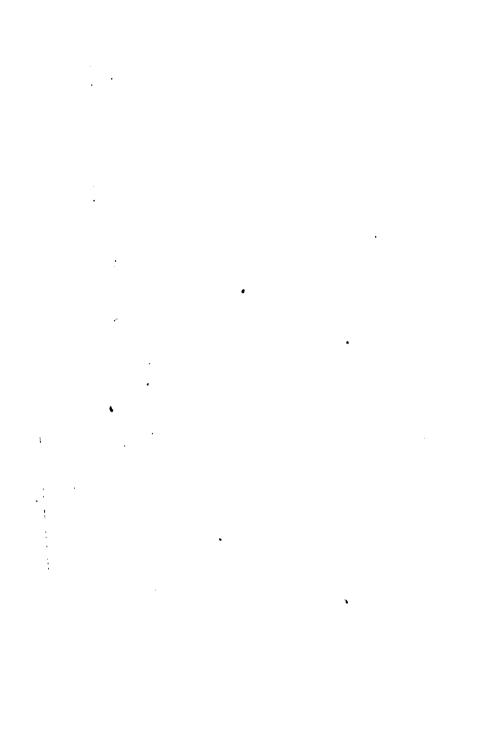
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# THE OWENS COLLEGE COURSE OF ELEMENTARY BIOLOGY.

PART I.

## THE FROG:

AN INTRODUCTION TO

# ANATOMY AND HISTOLOGY.

BY

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J. E. CORNISH, 33 PICCADILLY.

LONDON:

SMITH, ELDER, & CO., WATERLOO PLACE.

1882.

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#### PREFACE.

THE Owens College Course of Elementary Biology, which forms part of the scheme of study prescribed by the Victoria University, is of a rather more extended and comprehensive nature than the courses held elsewhere under the same name; and experience has shown me that there is want of a book that will guide and direct the student through the practical part of his work, the whole ground of which is covered by no one of the existing manuals. It is to meet this want that the present little work has been prepared.

This first instalment of the work consists of an Introduction containing practical instruction in the methods employed in biological investigation; followed by the application of these methods to the examination, both anatomical and histological, of an actual animal. For this purpose the frog has been selected as being convenient to dissect, easy to obtain, and a fairly typical example of the great group of Vertebrate animals. Where, from its small size or for other reason, the frog proved unsuitable, other animals have been substituted for it.

For convenience of reference, and in order to definitely stamp the practical character of the work, directions for dissection, etc., have throughout been printed in italics.

It is not expected that the student should do the whole of the work here given the first time he goes over it. The dissection of the muscles and of the cranial nerves should only be attempted if time remain after the other work is completed.

The second part of the book, which will include directions for the examination and dissection of certain animals selected as types of the leading animal groups, will, it is hoped, be ready before Christmas. In preparing this first part I have received very valuable assistance from Dr. Hartog, Demonstrator of Biology in the College, and from my friend and pupil Mr. C. H. Hurst. I am also much indebted to Prof. Gamgee and to Mr. Waters for the important help they have given me in the Histological portions.

OWENS COLLEGE,

August, 1882.

## INTRODUCTION.

#### I.-LABORATORY RULES.

1. The Laboratory is open daily throughout the session from 10 to 1.30 and from 2 to 5; on Saturdays from 10 to 1.

On Tuesdays and Thursdays the Laboratory is reserved for students taking the Elementary Biology course, who are

required to attend for three hours on each day.

3. Each student has a definite seat assigned him in the Laboratory, which he is not allowed to change without permission.

4. Each student has the free use of the drawer belonging to his seat: the key may be obtained on payment of a deposit of half a crown, which will be returned if the key is given up before the end of the term, but otherwise will be forfeited.

5. Each student is provided with a microscope, for the proper

treatment of which he is held responsible.

6. All necessary reagents and specimens for dissection are provided by the Laboratory, but each student is required to furnish himself with dissecting instruments, note book, and pencil, as explained in the next section.

Paper and solid refuse from dissections must not be thrown into the sinks, but into the pails provided for the purpose.

#### II.-LIST OF APPARATUS REQUIRED.

Each student is required to provide himself with the following:-

1. Two scalpels or dissecting knives, one large and one small.

2. Two pairs of forceps, one large and one small. Both pairs should have the tips serrated in order to secure a firmer hold.

- 3. Two pairs of scissors; one pair large and strong, for cutting bone and other hard tissues; the other pair small, for fine dissections. The latter pair should have the blades either bent at an angle (elbow scissors), or else curved. In selecting scissors be careful to see that they cut quite up to the points of the blades.
- 4. Three needles, mounted in handles; two round and one triangular.

5. A seeker, i.e. a blunt needle mounted in a handle, and with the end bent at a right angle.

6. A blowpipe, either of glass or metal.

- A razor, together with some means of keeping it sharp.
   Slides and coverslips, for mounting microscopical specimens.
- 9. A blank note book, for drawing in; an HB pencil, and a piece of india-rubber.

#### III.-ON DISSECTION.

The object of dissection is to separate the several parts and organs from one another, so as to define their boundaries and display clearly their relations to one another. Dissection consists, therefore, mainly in removing the "connective tissue" which binds the several parts to one another.

The following rules should be carefully attended to:-

1. Always pin down the animal you are dissecting firmly to the dissecting board. Never attempt to dissect a specimen that is not so fixed.

2. Never cut away anything until you are quite certain what

it is you are removing.

3. Fine dissections should always be done under water, which supports the parts and greatly facilitates the operation. A stream of water allowed to play gently on the dissection from time to time is often a valuable aid.

4. In pinning out a dissection stick the pins in, not vertically, but obliquely, so that their heads do not get in the way

or obscure the dissection.

5. Always put the part you are dissecting slightly on the stretch; e.g. when dissecting the bloodvessels or nerves of the throat, distend it by passing a small roll of paper or the handle of a seeker down the cosophagus; or when dissecting the muscles of the leg, pin out the leg in such a position as to stretch the muscles you are cleaning.

6. In cleaning bloodvessels or nerves always dissect along them and not across them; and avoid laying hold of them with the forceps. Similarly when cleaning muscles, dissect

along their fibres and not across them.

The dissection of muscles, and still more of nerves, is greatly aided by placing the specimens in spirit for a day before dissecting. 8. Always keep your instruments clean and sharp. Be careful not to blunt your fine scissors or scalpel by using them for cutting hard parts.

#### IV .- ON DRAWING.

It is absolutely essential to draw your dissections, and this must on no account be omitted. Keep a separate book for your drawings, and draw every dissection you make. Do not be discouraged if you find it difficult at first: you will never regret time spent on it.

The following rules will be useful to those who have not learnt

drawing systematically:

1. Always make your drawing to scale, i.e., either the exact size of the natural object, or half or double or treble that size as the case may be.

2. In commencing a drawing, first determine by careful measurement the positions of the principal points, and sketch in lightly the whole outline before finishing any one part.

3. If the object you are drawing is bilaterally symmetrical, draw a faint line down the middle of your paper, and sketch in the left hand half first; by measuring from your median line it will be very easy to make the two halves symmetrical.

4. Always name the several parts shown in your drawing, and mark also the scale adopted. Thus, if your drawing be of the natural size mark it thus— $\times 1$ ; if it be double the size of the object mark it  $\times 2$ ; if half the size,  $\times \frac{1}{2}$ , and so on.

5. Coloured pencils are very useful, and water-colour paints still better. If you use colours at all always keep certain colours for certain organs or tissues; e.g., when drawing the skeleton always colour the cartilage blue, the cartilage bones yellow, and the membrane bones either red or white; when drawing the bloodvessels colour the arteries red and the veins blue.

#### V.-THE USE OF THE MICROSCOPE.

The microscope consists essentially of a *stand* and a *body*, the latter of which bears at each end the *lenses* by which the magnifying power is obtained.

The stand is an upright pillar, the lower end of which is attached to a heavy foot to ensure steadiness. A little way above the foot the stand supports a horizontal plate—the stage

—on which the object to be examined is placed. The stage is perforated in the middle by a hole, the size of which can be varied by means of diaphragm. Through this hole light is directed on to the object to be examined by means of a mirror attached to the stand below the stage. Above the stage the stand supports a vertical tube, in which slides up and down the body of the microscope.

The body is a tube, in the upper end of which is placed a combination of lenses, known as the eyepiece, while to the lower end is screwed another combination of lenses—the objective.

Each microscope is provided with a couple of eyepieces and a couple of objectives of different magnifying power. An objective magnifying only a small number of times is called a low power; one magnifying many times (200 diameters or more) a high power. Similarly eyepieces are spoken of as high or low

according to their magnifying power.

In order that an object may be seen clearly the objective must be at a certain definite distance from the object, the distance varying with different objectives, and to a slight extent with different observers. The higher the power employed the closer must the objective be brought to the object. As the position of the object on the stage of the microscope is a fixed one this distance is regulated by moving the body of the microscope up and down in the tube in which it slides.

This process of focusing is effected in two ways: (1) by simply sliding the body up and down by hand: this is known as the coarse adjustment; it should always be performed with a slight screwing motion, and can only be used when low powers

are being employed.

(2) With high powers the distance between the objective and the object examined is so small that a more delicate method of adjustment is necessary. This fine adjustment is effected by a screw with a milled head placed at the top of the vertical pillar forming the stand. By turning the head from left to right, in the direction of an ordinary screw, the body of the microscope is lowered and the objective brought nearer to the object: by turning in the reverse direction the objective is raised.

In using the microscope attend to the following rules:-

1. Always examine an object first with the low power. Having adjusted the eyepiece and objective, direct the light up the

tube of the microscope by means of the mirror, and then place the object on the stage. Twist down the body until the objective is about a quarter of an inch from the object; look down the microscope, and gradually twist the body up until the object becomes visible. Focus accurately by means of the fine

adjustment.

2. When using a high power, start with the objective the same distance from the object as before, and very gradually twist down the body until the object becomes dimly visible; then complete the focusing by the fine adjustment. It will facilitate the process if, while twisting down the body with the right hand, you move about the object slightly with the left hand.

3. Take extreme care never to let the objective touch the object; and never to touch or allow any dirt to get on the face of the objective. The face of an objective cannot be cleaned

without doing harm to it.

4. Should by any chance a drop of glycerine get on the face of the objective, stream it gently with water from a washbottle, and wipe very carefully with a silk handkerchief or piece of chamois leather. Should Canada balsam be allowed to get on the objective, do not attempt to clean it yourself, but give it at once to the assistant.

 See that the body of the microscope slides smoothly in its tube. If it does not, remove it, and clean it by rubbing with

a few drops of olive oil.

6. Keep the eye you are not using open; a very little practice will enable you to do this, and it will save you much fatigue.

Also get in the habit of using either eye.

7. With a high power, use a small diaphragm: the amount of light will be somewhat diminished, but the clearness and definition of the object much increased.

8. When examining an object, use your left hand to move the object about on the stage, and keep your right hand on the

fine adjustment.

9. If the object appears dim or dirty, find out where the

fault lies, in this way :-

While looking down the microscope, turn round the eyepiece with your right hand: if the dirt turns round too, remove and clean the eyepiece. If the fault is not in the eyepiece, move the slide about gently; if the dirt moves with the slide remove

the slide and clean it. If the dirt does not move with either the eyepiece or the slide the fault is almost certainly in the objective, which should be removed and examined; if dirty, it must be cleaned very carefully with a piece of silk or chamois leather.

## VI.-THE PREPARATION OF MICROSCOPIC OBJECTS.

In mounting microscopic objects be careful that your slides and coverslips are thoroughly clean. Slides should be labelled as soon as they are prepared, and should be kept in a box or cabinet in which they can lie flat.

## A. Methods of Mounting.

There are various media in which objects may be mounted. The method of procedure is much the same with all. Put a small drop of the fluid in the middle of the slide, place the object in the middle of the drop, and arrange it with needles in any position that may be desired. Then place the coverglass carefully on the top, letting it rest by one edge on the slide and supporting the opposite edge by a needle, which is gradually withdrawn so as to let the coverglass down slowly and drive out any air-bubbles there may be in the fluid. If any air-bubbles still remain, leave them alone, as they will probably work out by themselves. Be careful not to use too large a drop of your mounting medium, and above all things be careful not to let any of it get on the top of the coverglass; should this happen, the cover must be removed at once and the specimen mounted again with a clean coverglass.

The most important mounting media are :-

- Water. Used for most vegetable tissues, but must not be employed for animal tissues. If the preparation appears cloudy it should be immersed in solution of caustic potash before mounting. If it is still obscured by bubbles, these may be removed by immersing the specimen in spirit until they disappear, and then mounting in water.
- 2. Normal Salt Solution. A 0.75 per cent. solution of common salt in water. This is very useful for the examination of fresh specimens of animal tissues, as, unlike water, it has practically no action whatever on them. It cannot be used, however for making permanent preparations.

- 3. Glycerine. Can be used either pure or diluted with its own bulk of water. If the specimens are intended to be permanent preparations, a narrow ring of cement must be painted round the edge of the coverglass to fix it to the slide, and to prevent evaporation.
- 4. Canada Balsam. The most generally useful; requires no cement. Specimens that are to be mounted in balsam must first be deprived of all water they may contain by placing for a day in absolute alcohol, and should then, before mounting, be soaked for a few minutes in a mixture of creosote and turpentine in order to clear them, i.e., render them transparent. Canada balsam, if too thick, may be diluted with chloroform, turpentine, or benzole.

## B. Teasing.

The object of teasing is to separate the several parts of a tissue or organ from one another as completely as possible, in

order to show their ultimate structure.

The fragment to be teased should be placed on a slide in a drop of the medium in which it is to be mounted and then torn up into as minute particles as possible by means of a couple of needles held one in each hand. The process is often greatly facilitated by placing the slide on a piece of black paper, which renders the particles easier to see. When torn up as finely as possible, a coverglass is placed on as before. The two rules to be borne in mind in teasing are—

1. Take a very small fragment to commence with.

2. Tease it as finely as you possibly can. Your object is to separate the component cells from one another.

#### C. Maceration.

The process of teasing is in many cases very greatly facilitated by previously macerating the specimen, i.e., soaking it in some fluid, which, while preserving the individual cells, tends to loosen their connections with one another. The most important macerating fluids are—

- 1. Ranvier's Alcohol. A mixture of one part of strong spirit with two parts of water. The specimens should be put fresh into this preparation and allowed to remain twenty-four hours.
- 2. Baryta Water. Very useful for isolating the individual cells of tendons.

3. Muller's Fluid. A solution of bichromate of potash with a little sodic sulphate in water.

## D. Staining.

Various reagents are employed for the purpose of staining preparations; some of these merely colour the whole preparation more or less uniformly; but the most useful ones are those which stain certain parts of the cells only, or at any rate stain these much more strongly than the other parts. The most important are:

- 1. Hæmatoxylin. There are various preparations of hæmatoxylin, or logwood, used in microscopical work: the best is that proposed by Kleinenberg and called by his name. The specimens, which must be perfectly free from all trace of acid, should be left in the hæmatoxylin in a covered vessel or stoppered bottle for from one to twenty-four hours, according to the depth of staining desired, and then placed in strong spirit for some hours before mounting. Hæmatoxylin stains the nuclei of cells much more strongly than the other parts.
- 2. Alum-carmine. A solution in water stains small pieces of tissues well, but will not penetrate larger masses. After staining, the specimen should be washed in water and mounted in glycerine, or else dehydrated by means of alcohol and mounted in balsam.
- 3. Picro-carmine. A very useful and, to a certain extent, a differential stain, as it stains different tissues slightly different colours. Answers best with specimens preserved in 70 per cent. alcohol: these should be placed in picro-carmine for a day, then washed with water and placed for an hour in 1 per cent. acetic acid: they are then placed in 70 per cent. alcohol, in which they are left so long as any colour is dissolved out by it, and then mounted.
- 4. Magenta. Stains very rapidly, but diffusely: the colour also is not permanent. Useful for vegetable tissues and for infusoria.
- 5. Iodine solution. Chiefly used with vegetable tissues for the detection of starch, which it stains a blue or violet colour.
- 6. Silver nitrate. A 1 per cent. solution in water stains the intercellular substance, which binds together the several cells of a tissue, much more strongly than the cells themselves, and is

therefore chiefly used when we wish to render prominent the outlines of the individual cells. The specimens should be placed fresh in the silver solution for from two minutes to a quarter of an hour, then washed thoroughly with distilled water and exposed to the light until stained sufficiently deeply, when they may be mounted in glycerine. Such preparations are rarely permanent, as the reduction of the silver, to which the staining is due, continues until the specimens become ultimately too dark to be of any use.

- 7. Osmic Acid. A 1 per cent. solution of osmic acid in water forms an extremely useful staining agent. It is especially useful for the detection of fat which is stained by it a dark brown or black colour. Specimens, which must be quite fresh, should only be left in it a few minutes, and may then be mounted in glycerine or else dehydrated and mounted in balsam.
- 8. Acetic Acid. Although not strictly a staining agent inasmuch as it does not colour the specimens, acetic acid may conveniently be mentioned here as it is used for the same purpose as the true stains, i.e., for the sake of rendering certain parts of the cells especially distinct. Acetic acid, of which a 1 per cent. solution is employed, causes the protoplasm of cells to swell up and become transparent, and brings into special prominence the nuclei. It is used with fresh specimens of both animal and vegetable tissues.

#### VII.-ON SECTION CUTTING.

In order to study many tissues and organs it is necessary to cut them into thin sections; and this method of investigation is of such importance as to require special notice. There are three chief stages: Hardening, Imbedding, and Cutting, which will be noticed in succession.

## A. Hardening.

Before the object can be cut into sections it is necessary to harden it: this may be effected by freezing, but the more usual plan is by means of reagents. The general action of these hardening reagents is to coagulate the protoplasm of the tissues; and the objects to be attained are to effect this coagulation quickly, before the tissues can undergo any alteration; and thoroughly, i.e., throughout the whole thickness of the object to be hardened. To ensure the latter result it is always advisable to use small pieces of the substance to be cut.

3. Muller's Fluid. A solution of bichromate of potash with a little sodic sulphate in water.

## D. Staining.

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plant, a convenient way of cutting it is to take a piece of carrot, make a vertical slit in it and insert the leaf in the slit; by cutting with a razor thin slices of the carrot and the leaf together, sections of the leaf of suitable thinness can readily be obtained.

In dealing with animal tissues such a process is inapplicable, owing to the danger of compressing the specimen; and in order to cut sections of such specimens it is usual to imbed them in a waxy substance.

There are various methods of doing this, differing chiefly in points of detail. The two most usual plans are the following:

1. The Spermaceti Method. Stain the specimen as described above. After removing it from the staining fluid place it in absolute alcohol for 24 hours in order to completely remove any water it may contain.

Then transfer it to oil of bergamot for about half an hour: the oil readily soaks through the specimen, and so enables the spermaceti at the next stage of the process to penetrate more

readily and completely.

In the meanwhile take a stiffish piece of paper 4½ inches long by 3 inches wide, and fold it so as to make a paper box an inch and a half long by an inch in width and depth. Make a mixture of four parts of spermaceti with one part of castor oil; melt it, half fill the paper box with the mixture, and allow it to cool.

Take the specimen out of the oil of bergamot, dry it slightly with blotting paper, and place it in a watch glass filled with the spermaceti mixture: keep this just melted for about half an hour in order to allow the spermaceti to thoroughly penetrate the

specimen.

When the spermaceti in the box has thoroughly set, pour the specimen from the watch glass into the box, and fill up the box with melted spermaceti. Place the specimen close to one end of the box and in the desired position, moving it with needles. Make marks on the outside of the box to remind you of the position of the specimen, and let the spermaceti cool, stirring out with a hot needle any bubbles that may form.

2. The Paraffin Method. In this method paraffin is used instead of the mixture of spermaceti and castor oil. The specimen is transferred from the essential oil to turpentine, in which it is left half an hour; it is then placed in paraffin which is kept just above its melting point for 24 hours, to let the

paraffin soak in thoroughly; it is then imbedded as before. In both methods great care must be taken not to overheat the specimen.

## E. Cutting.

1. The Spermaceti Method. When the block has thoroughly set, which takes some hours, remove it from the box and cut away the spermaceti with a knife until the specimen just comes into view. Then cut sections one by one with a razor, either holding the block in your hand or else placing it in a microtome. Keep the razor wetted with olive oil, and float off each section as it is made on to a glass slide. Arrange the sections in order on the slide with a needle, and remove the superfluous oil with a cloth or with blotting paper, to prevent the sections floating about and so getting out of position.

When you have got as many sections on the slide as will go

under one coverglass, begin another slide.

When all the sections are cut, clear them, i.e. remove the oil and spermaceti, by means of a mixture of creosote and turpentine. Place a few drops of the mixture along one side of the sections and note how the spermaceti is dissolved, and together with the oil driven over to the opposite side of the slide: carefully sop it up with a cloth or with blotting paper: replace with a needle any sections that may have moved from their places, and repeat the process until the sections are perfectly clear, and when examined under the microscope show no crystals of spermaceti.

When quite clear dry up the creosote and turpentine, or allow it to evaporate, and then apply a small quantity of Canada balsam, just enough to cover the sections and fix them in their places: leave the slide in this condition until the next day, when the sections will be firmly fixed: then place a small drop of balsam on the top, and put on the coverglass. Label the slide, and mark on the label, by numbers, the order in which

the successive sections are arranged.

2. The Paraffin Method. Specimens imbedded in paraffin are cut in a very similar manner. Before commencing the sections paint the slide with a solution of shellac in creosote. Cut with the razor dry, and transfer each section by a camel hair brush to its proper place on the slide, where it will adhere the shellac and creosote. When all the sections are cut,

warm the slide so as to melt the paraffin; then wash it thoroughly with turpentine and mount in balsam.

The great advantage of this method is that there is no danger of the sections shifting their position on the slide, and therefore much time is saved that in the first method is devoted to watching and replacing the sections which float from their proper places.

## Chap. I.-GENERAL ANATOMY OF THE FROG.

#### A. External Characters.

Lay the frog on a board before you, and note the following points:—

- 1. The division into head, trunk, and limbs; and the absence of neck and tail.
  - 2. The two great surfaces :
    - a. The dorsal surface, or back, directed upwards when the frog is in the natural position.
    - b. The ventral surface, or belly, directed downwards towards the ground.
- 3. The skin: moist and smooth: devoid of hairs, scales, or claws. The colour of the skin is variable in different specimens and at different times: mottled on the dorsal surface, paler on the ventral.
- 4. The head: flat and triangular, with a blunt apex directed forwards.

At the sides of the head are the eyes—large and prominent. Each eye has two eyelids of which the upper is thick, pigmented, and almost immovable, while the lower is semi-transparent and freely movable.

Behind the eye on either side is an obliquely placed elongated patch of a dark colour, in the middle of which is a circular area—the tympanic membrane—supported by a firm marginal ring.

- 5. The limbs: two pairs, fore and hind, each composed of three segments
  - a. Fore limb:
    - i. Arm.
    - ii. Forearm.
    - iii. Hand, with four digits corresponding to the four fingers of man; the thumb being unrepresented. In the male frog at the breeding season there is a thickening along the inner edge of the first digit.

- b. Hind limb: much longer than the fore limb: composed of
  - i. Thigh. ii. Leg.
  - iii. Foot, with five toes, webbed together. The shortest toe corresponds to the big toe of man, and the longest to his fourth toe.
- 6. External apertures: or openings on the surface of the body.
  - a. Median apertures.
    - i. Mouth: a wide horizontal slit.
    - ii. Cloacal aperture: a small hole at the posterior end of the body between the legs: placed slightly on the dorsal surface, just behind the bony projection formed by the posterior end of the urostyle.
  - b. Paired apertures.
    - Nostrils or anterior nares: two small openings on the dorsal surface of the head, close to its anterior end.

## B. The Buccal Cavity.

Open the mouth to its full extent: note the wide buccal or mouth cavity, of which the hinder part or pharynx is continued back into the esophagus. Note also the following structures:—

- 1. On the Roof of the Mouth.
  - a. Teeth.

i. Maxillary teeth: a row of fine teeth attached

round the edge of the upper jaw.

ii. Vomerine teeth: two small patches of sharp teeth in the fore part of the roof of the mouth and near the middle line.

b. The posterior nares. Two small holes lying to the outer sides of and slightly in front of the two patches of vomerine teeth.

Pass bristles through the nostrils, and see that they come out through the posterior nares into the buccal

cavity.

c. The Eustachian tubes or recesses. A pair of much larger holes at the sides of the posterior part of the buccal cavity. Each hole opens into a slightly dilated chamber—the tympanic cavity—which is closed externally by the tympanic membrane already seen on the surface of the head.

Perforate the tympanic membrane on one side with a needle, and pass a bristle through the hole and down the

Eustachian tube into the mouth.

d. Two rounded prominences at the sides of the roof of

the mouth caused by the eyeballs.

Press down one of the eyes with your finger, and note that it can be made to project very considerably into the buccal cavity.

#### 2. On the Floor of the Mouth.

- a. The lower jaw, devoid of teeth, and forming a bony margin to the floor of the mouth: the rest of the floor is soft and fleshy, but is slightly stiffened by a cartilaginous plate—the body of the hyoid.
- b. The tongue: thin and fleshy: attached to the front part of the floor of the mouth, and with its free bilobed end turned backwards toward the throat.

  Turn the tongue forward with the forceps to see
- c. The glottis: a small longitudinal slit on the floor of the posterior part of the mouth: its sides are stiffened by the arytenoid cartilages.

Pass bristles through the glottis into the lungs.

### C. The Abdominal Viscera.

Lay the frog on its back under water, and fasten it down to the dissecting board by pins through the limbs. Cut through the skin along the middle line the whole length of the ventral surface. Separate the skin from the underlying parts, noticing its very loose attachment to these parts, and the large space—lymph cavity—beneath it. Turn the flaps of skin out and pin them back. Notice

a. The muscles forming the body wall.

b. The sternum, or "breast-bone," in the middle line, opposite the fore-limbs. Pinch up with forceps the muscular body wall, and cut through it with scissors a little to the right of the median line into the body cavity, being careful not to injure the anterior abdominal vein which runs along the inner surface of the body wall in the middle line.

Continue the cut backwards to the hinder end of the body and forwards to the jaw, cutting through the sternum with strong

scissors, and taking care not to injure the parts beneath.

Note on the inner surface of the left flap the anterior abdominal vein, and carefully dissect this from the flap. Pull the two flaps apart, cutting through them transversely at their posterior ends to facilitate the process, and pin them out so as to display the viscera.

Inflate the lungs with a blowpipe through the glottis, and inflate

the bladder through the cloacal aperture.

Note and draw the general arrangement of the viscera, showing:—

- 1. The heart: enclosed in the pericardium: situated in the middle line in front, and in the natural condition of the parts covered by the sternum.
- 2. The lungs: two thin-walled elastic sacs at the sides of the heart.

Note the bristles already passed into the lungs through the glottis.

- 3. The liver: a large reddish brown bilobed organ just behind the heart.
- 4. The corpora adiposa, or fat bodies: two bright yellow tufts of flattened processes attached to the dorsal wall of the body cavity: they vary much in size, and usually come to the surface just behind the liver.
- 5. The intestine: a light coloured convoluted tube: in the middle line behind is the much wider large intestine.
- 6. The bladder: a thin walled bilobed sac at the posterior end of the body cavity.
  - 7. In the female frog note, in addition to the above parts,
    - a. The ovaries: two large bodies of irregular shape, each consisting of a mass of spherical black and white eggs, like small shot.
    - b. The oviducts: two long very much convoluted tubes with thick white walls, lying at the sides of the body cavity.

3. Muller's Fluid. A solution of bichromate of potash with a little sodic sulphate in water.

## D. Staining.

Various reagents are employed for the purpose of staining preparations; some of these merely colour the whole preparation more or less uniformly; but the most useful ones are those which stain certain parts of the cells only, or at any rate stain these much more strongly than the other parts. The most important are:

- 1. Hæmatoxylin. There are various preparations of hæmatoxylin, or logwood, used in microscopical work: the best is that proposed by Kleinenberg and called by his name. The specimens, which must be perfectly free from all trace of acid, should be left in the hæmatoxylin in a covered vessel or stoppered bottle for from one to twenty-four hours, according to the depth of staining desired, and then placed in strong spirit for some hours before mounting. Hæmatoxylin stains the nuclei of cells much more strongly than the other parts.
- 2. Alum-carmine. A solution in water stains small pieces of tissues well, but will not penetrate larger masses. After staining, the specimen should be washed in water and mounted in glycerine, or else dehydrated by means of alcohol and mounted in balsam.
- 3. Picro-carmine. A very useful and, to a certain extent, a differential stain, as it stains different tissues slightly different colours. Answers best with specimens preserved in 70 per cent. alcohol: these should be placed in picro-carmine for a day, then washed with water and placed for an hour in 1 per cent. acetic acid: they are then placed in 70 per cent. alcohol, in which they are left so long as any colour is dissolved out by it, and then mounted.
- 4. Magenta. Stains very rapidly, but diffusely: the colour also is not permanent. Useful for vegetable tissues and for infusoria.
- 5. Iodine solution. Chiefly used with vegetable tissues for the detection of starch, which it stains a blue or violet colour.
- 6. Silver nitrate. A 1 per cent. solution in water stains the intercellular substance, which binds together the several cells of a tissue, much more strongly than the cells themselves, and is

therefore chiefly used when we wish to render prominent the outlines of the individual cells. The specimens should be placed fresh in the silver solution for from two minutes to a quarter of an hour, then washed thoroughly with distilled water and exposed to the light until stained sufficiently deeply, when they may be mounted in glycerine. Such preparations are rarely permanent, as the reduction of the silver, to which the staining is due, continues until the specimens become ultimately too dark to be of any use.

- 7. Osmic Acid. A 1 per cent. solution of osmic acid in water forms an extremely useful staining agent. It is especially useful for the detection of fat which is stained by it a dark brown or black colour. Specimens, which must be quite fresh, should only be left in it a few minutes, and may then be mounted in glycerine or else dehydrated and mounted in balsam.
- 8. Acetic Acid. Although not strictly a staining agent inasmuch as it does not colour the specimens, acetic acid may conveniently be mentioned here as it is used for the same purpose as the true stains, i.e., for the sake of rendering certain parts of the cells especially distinct. Acetic acid, of which a 1 per cent. solution is employed, causes the protoplasm of cells to swell up and become transparent, and brings into special prominence the nuclei. It is used with fresh specimens of both animal and vegetable tissues.

## VII.-ON SECTION CUTTING.

In order to study many tissues and organs it is necessary to cut them into thin sections; and this method of investigation is of such importance as to require special notice. There are three chief stages: Hardening, Imbedding, and Cutting, which will be noticed in succession.

## A. Hardening.

Before the object can be cut into sections it is necessary to harden it: this may be effected by freezing, but the more usual plan is by means of reagents. The general action of these hardening reagents is to coagulate the protoplasm of the tissues; and the objects to be attained are to effect this coagulation quickly, before the tissues can undergo any alteration; and thoroughly, i.e., throughout the whole thickness of the object to be hardened. To ensure the latter result it is always advisable to use small pieces of the substance to be cut.

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The hardening reagents in most common use are:

- 1. Osmic Acid. For this purpose a 1 per cent. solution in water is used: it acts almost instantaneously, and so allows no change to occur in the tissues; it has also the merit of staining the tissues as well as hardening them. It can, however, only be employed when the specimens are very small, as it hardens the surface layers so rapidly that it is unable to penetrate beyond a very slight depth. A few minutes immersion is usually sufficient.
- 2. Chromic Acid. A 0.25 to 0.5 per cent. solution of chromic acid in water is a useful hardening agent; it acts much more slowly than osmic acid, but is thereby enabled to penetrate to greater depths. Specimens should usually be left in the solution for 24 hours.
- 3. A Mixture of chromic acid with a few drops of osmic acid is often very useful, as it combines the advantages of both reagents.
- 4. Picric Acid. A very valuable hardening reagent, of which the best preparation is Kleinenberg's. Specimens should be left in it from 12 to 24 hours.
  - 5. Absolute Alcohol. Often a useful hardening reagent.

## B. Dehydration.

With the exception of the last, specimens that have been hardened in any of the preceding reagents should, on removal from them, be placed for a day in 30 per cent, alcohol, and then gradually dehydrated by transferring them every day to alcohol of greater strength. Thus on the second day they should be transferred to 50 per cent, alcohol, on the third day to 70 per cent, and on the fourth day to 90 per cent, in which they may be left till required.

## C. Staining.

The hardened specimens, if not too large, may now be stained with either hæmatoxylin or piero-carmine; they should then be placed back in 90 per cent. alcohol and transferred from that to absolute alcohol. If too large to stain whole, the sections must be stained separately after cutting.

## D. Imbedding.

If the object to be cut be flat and thin, like the leaf of a

plant, a convenient way of cutting it is to take a piece of carrot, make a vertical slit in it and insert the leaf in the slit; by cutting with a razor thin slices of the carrot and the leaf together, sections of the leaf of suitable thinness can readily be obtained.

In dealing with animal tissues such a process is inapplicable, owing to the danger of compressing the specimen; and in order to cut sections of such specimens it is usual to imbed them in

a waxy substance.

There are various methods of doing this, differing chiefly in points of detail. The two most usual plans are the following:

1. The Spermaceti Method. Stain the specimen as described above. After removing it from the staining fluid place it in absolute alcohol for 24 hours in order to completely remove any water it may contain.

Then transfer it to oil of bergamot for about half an hour: the oil readily soaks through the specimen, and so enables the spermaceti at the next stage of the process to penetrate more

readily and completely.

In the meanwhile take a stiffish piece of paper 4½ inches long by 3 inches wide, and fold it so as to make a paper box an inch and a half long by an inch in width and depth. Make a mixture of four parts of spermaceti with one part of castor oil; melt it, half fill the paper box with the mixture, and allow it to cool.

Take the specimen out of the oil of bergamot, dry it slightly with blotting paper, and place it in a watch glass filled with the spermaceti mixture: keep this just melted for about half an hour in order to allow the spermaceti to thoroughly penetrate the

specimen.

When the spermaceti in the box has thoroughly set, pour the specimen from the watch glass into the box, and fill up the box with melted spermaceti. Place the specimen close to one end of the box and in the desired position, moving it with needles. Make marks on the outside of the box to remind you of the position of the specimen, and let the spermaceti cool, stirring out with a hot needle any bubbles that may form.

2. The Paraffin Method. In this method paraffin is used instead of the mixture of spermaceti and castor oil. The specimen is transferred from the essential oil to turpentine, in which it is left half an hour; it is then placed in paraffin which is kept just above its melting point for 24 hours, to let the

#### b. Cells in situ:

Snip off a small piece of the epithelium from the tongue of a freshly killed frog: mount in normal salt solution and add a small drop of gamboge water to render the movements more visible: examine with high power; note

GLANDS.

- i. The currents due to the ciliary motion.
- ii. The movements of the individual cilia: best seen when the specimen is beginning to die, and the movements to slacken in speed.
- 4. Stratified epithelium: cells arranged in several layers. Take prepared section of asophagus of rabbit, or of conjunctiva of rabbit or pig, which has been hardened in chromic acid, stained, and cleared in creosote and turpentine. Mount in balsam, examine with high power, and note
  - i. The stratification of the epithelium.
  - ii. The transition from the deeper spherical or columnar cells to the superficial squamous cells.

#### B. Glands.

A gland consists essentially of a layer of epithelial cells secreting some special fluid. The epithelial surface may be flat, but is more usually folded, often in a very complicated manner, in order to increase the extent of the secreting surface.

a. Simple Glands. In simple glands the epithelial surface is increased by simple undivided pit-like depressions, whose mouths serve to discharge the secretion on the free surface.

Take prepared section of large intestine of rabbit which has been hardened in chromic acid, stained, and cleared in creosote and turpentine. Mount in balsam, and examine first with low power, then with high. Note

- The shape of the glands: simple pit-like depressions of the surface.
- ii. The characters of the glandular epithelium lining the pits: a single layer of short columnar granular cells.

#### b. Cells in situ: cast skin of newt.

Take a small piece of the prepared specimen, which has been stained in hamatoxylin, and then, after treatment with alcohol, cleared with creosote and turpentine. Mount the specimen in balsam; cover and examine with a high power: note

- i. Flattened cells fitted together at their edges to form a continuous layer: each cell has a large nucleus near its centre.
- 2. Columnar epithelium: elongated rodlike cells placed vertically to the surface on which they rest. If the epithelium is stratified the columnar character is most marked in the most superficial cells.
  - a. Isolated cells. From small intestine of frog: isolated by maceration for 24 hours in Ranvier's alcohol, and stained with piero-carmine.

Mount a small drop of the prepared specimen in glycerine: paint a ring of cement round the coverglass to prevent evaporation; and examine with high power: note:—

 i. Columnar shape of the cells: nuclei. The cells often remain side by side in little groups.

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Take prepared section of stomach of dog which has been stained and then cleared in creosote and turpentine. Mount in balsam and examine with high power: note

- Superficial layer of long narrow columnar cells packed together side by side; with nuclei at their inner or deeper ends.
- 3. Ciliated epithelium: the cells, which are usually columnar, bear at their free ends tufts of exceedingly fine hair-like processes—cilia—which, when living, exhibit active lashing movements.
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 Columnar shape of the cells: nuclei. The cells often remain side by side in little groups.

#### b. Cells in situ.

Take prepared section of stomach of dog which has been stained and then cleared in creosote and turpentine. Mount in balsam and examine with high power: note

- Superficial layer of long narrow columnar cells packed together side by side; with nuclei at their inner or deeper ends.
- 3. Ciliated epithelium: the cells, which are usually columnar, bear at their free ends tufts of exceedingly fine hair-like processes—cilia—which, when living, exhibit active lashing movements.
  - a. Isolated cells. From trachea of rabbit: isolated by maceration for 24 hours in Ranvier's alcohol; stained with picro-carmine, and scraped into glycerine.

Mount a small drop of the prepared specimen in glycerine: paint a ring of cement round the coverglass: examine with high power, and note

i. The shape of the cells; their nuclei; and the tuft of cilia at one end.

#### b. Cells in situ:

Snip off a small piece of the epithelium from the tongue of a freshly killed frog: mount in normal salt solution and add a small drop of gamboge water to render the movements more visible: examine with high power; note

- i. The currents due to the ciliary motion.
- ii. The movements of the individual cilia: best seen when the specimen is beginning to die, and the movements to slacken in speed.
- 4. Stratified epithelium: cells arranged in several layers. Take prepared section of æsophagus of rabbit, or of conjunctiva of rabbit or pig, which has been hardened in chromic acid, stained, and cleared in creosote and turpentine. Mount in balsam, examine with high power, and note
  - i. The stratification of the epithelium.
  - ii. The transition from the deeper spherical or columnar cells to the superficial squamous cells.

### B. Glands.

A gland consists essentially of a layer of epithelial cells secreting some special fluid. The epithelial surface may be flat, but is more usually folded, often in a very complicated manner, in order to increase the extent of the secreting surface.

a. Simple Glands. In simple glands the epithelial surface is increased by simple undivided pit-like depressions, whose mouths serve to discharge the secretion on the free surface.

Take prepared section of large intestine of rabbit which has been hardened in chromic acid, stained, and cleared in crossote and turpentine. Mount in balsam, and examine first with low power, then with high. Note

- The shape of the glands: simple pit-like depressions of the surface.
- ii. The characters of the glandular epithelium lining the pits: a single layer of short columnar granular cells.

- Add a drop of acetic acid to the preparation: note that
  iii. The fibres swell up and become transparent.
  - iv. Rows of connective tissue corpuscles with nuclei become visible between the fibres.
  - b. Isolated fibres and fibrillæ: take a small piece of tendon that has been macerated for 24 hours in baryta water or picric acid to dissolve the cementing ground substance: tease it in a drop of glycerine: cover and examine with high power: note
    - i. Isolated fibres and fibrillæ.
- 2. Yellow elastic tissue: consists of fine branched homogeneous fibres, with great power of resisting chemical reagents: the fibres are formed from branched cells which lose their nuclei completely during development.
  - a. Ligamentum nuchæ of ox: tease finely a small shred in water: examine with low and high powers: note

i. The branching fibres, with very sharp outlines.

- The tendency of the branches to anastomose with one another and so form networks.
- The tendency of the fibres and branches to curl up at their broken ends.

Add a drop of acetic acid: note

- iv. No alteration whatever is produced in the fibres.
- v. No nuclei appear.
- Areolar tissue: a meshwork composed of both white fibrous and elastic tissues.
  - a. Subcutaneous tissue of mammal. Snip off a small piece from beneath the skin: spread on a slide: add a drop of normal salt solution: cover, and examine with low and high powers: note
    - Meshwork composed of white fibrous tissue with wavy outlines, mingled with which are branched elastic fibres.

Add acetic acid: note

- The white fibrous tissue swells up and becomes transparent.
- iii. The elastic tissue is unaltered.
- Connective tissue corpuscles with nuclei become evident.

- Adipose tissue: consists of a network of vascular connective tissue, in the meshes of which are fat cells, i.e., connective tissue corpuscles in which large quantities of fatty matter have accumulated.
  - a. Omentum of rabbit or kitten: mount a small piece of fresh omentum in normal salt solution; examine with low and high powers; note
    - i. Vascular connective tissue meshwork, in which lie
    - ii. Fat cells: large spherical, or from mutual pressure polygonal cells: distended with fatty matter, and with nucleus at one side.

## b. Osmic acid specimen.

Note the reduction of the osmic acid by the fat, which becomes stained a dark brown or black colour.

## E. Cartilage.

In cartilage or gristle the intercellular substance, which in most other tissues is only present in small quantity, is greatly increased so as in some cases to far exceed in bulk the cells which it connects together. The intercellular substance forms a dense translucent matrix resembling an extremely stiff jelly, in which are imbedded the cartilage cells, either singly or in groups. In young cartilage the intercellular substance is much less abundant, and the cells consequently closer together than they are in older or more mature forms.

- 1. Hyaline cartilage. Take a small piece of cartilage from the shoulder girdle of a newt: scrape away gently any muscle or other tissue that may adhere to it: mount in normal salt solution, and examine with low and high powers: note
  - The intercellular matrix: hyaline or faintly granular.
  - ii. The cartilage cells: imbedded in the matrix:
    each cell nucleated, and occupying a cavity or
    lacuna in the matrix. In places the cells are in
    groups of twos or fours from recent division.

Irrigate the specimen with acetic acid: examine again with high power, and note

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iii. The cells shrink away from the walls of the cavities in which they lie, and the nuclei become more distinct.

Wash the specimen thoroughly in water to remove the acid: stain with hæmatoxylin, and mount as a permanent preparation in glycerine: examine with high power, and note

- iv. The cell nuclei are stained deeply, and the matrix very slightly: the layer of matrix immediately surrounding each cell—the capsule—stains deeper than the other parts.
- 2. Articular cartilage. Forms caps covering the ends of those bones which fit together to form movable joints: these caps act as elastic cushions to break the force of shocks.

Mount in glycerine prepared section of articular cartilage from head of femur, the section being made perpendicular to the articular surface: examine with low and high powers: note

- i. The matrix: hyaline or faintly granular.
- ii. The cartilage cells: arranged in more or less parallel rows. Towards the free surface the cells become gradually flattened.

## F. Bone.

Bone consists of a dense fibrillar intercellular matrix, in which are imbedded cells which lie in cavities connected with one another by fine branching canals. The matrix is richly impregnated with inorganic salts, chiefly phosphate and carbonate of lime, which form about two-thirds by weight of the substance of the bone and gives it its great hardness and strength. The matrix, with its contained cells, is arranged in concentric layers, formed in succession one within another around central canals in which lie the bloodvessels, which penetrate the bone in great numbers. A bloodvessel with its surrounding layers of matrix and cells is spoken of as a **Haversian system**.

- 1. Examine with both low and high powers prepared transverse sections of a long bone: note
  - The Haversian systems: concentric layers surrounding the Haversian canals.

- ii. Incomplete Haversian systems: filling up the spaces between the complete systems. These are the remains of systems which were formerly complete but have been removed in part by the process of absorption which is continually going on in bone.
- iii. The lacunæ: or spaces in the matrix in which the bone cells lie.
- iv. The canaliculi: a network of very fine branching canals connecting the lacunæ together: probably occupied while the bone is living by branching processes of the bone-cells.
- v. The large central medullary cavity of the bone: occupied during life by the marrow.
- vi. The peripheral or circumferential lamellæ: forming the most superficial layer of the bone: a series of concentric lamellæ parallel to the surface.
- vii. The perimedullary lamellæ: a series of concentric lamellæ lining the central medullary cavity of the bone.

## Chap. IV.—THE SKELETON OF THE FROG.

The skeleton, which forms the hard internal parts of the frog, is composed partly of cartilage and partly of bone. It forms a framework giving definite shape to the body, and precision to the movements; and serves also to protect from injury some of the more important and delicate organs, notably the central nervous system, the sense organs, and the heart. In the early stages of development the skeleton consists entirely of cartilage: in the adult this primary cartilaginous skeleton is replaced to a greater or less extent in different parts by bone. Bone may also be developed in places where there was no pre-existing cartilage, and is then called membrane bone, in contradistinction to the former kind, or cartilage bone, which replaces pre-existing cartilage. Cartilage may also be calcified, i.e., have calcareous salts deposited in its matrix, without in any way taking on the character of true bone.

The skeleton may be conveniently divided into (1) the axial portion, including the skull and the vertebral column: and (2) the appendicular portion, including the limbs and the limb-girdles which attach them to the body.

Examine the prepared skeletons and make careful drawings to scale of the several parts. Colour, in your drawings, the cartilage blue, the cartilage bones yellow and the membrane bones white or red. Prepare skeletons for yourself by soaking the parts in hot water, and carefully brushing away the soft tissues until the skeleton is clean.

#### A. The Axial Skeleton.

I. The vertebral column or "back bone." Divisible into an anterior segmented portion, composed of nine vertebræ placed end to end, and a posterior unsegmented portion of nearly equal length—the urostyle. The whole vertebral column forms a bony tube, within which lies the spinal cord; and at the sides of the tube between the successive vertebræ are the intervertebral foramina through which the nerves pass out from the spinal cord to the various parts of the body.

- a. Examine one of the vertebræ, say the third, more closely: draw it, showing:
  - i. Its ring-like shape.
  - ii. The centrum or body: the thickened ventral portion of the ring: articulates closely with the centra of the vertebræ in front of and behind it; and forms the floor of the neural canal in which the spinal cord lies.
  - iii. The neural arch: the lateral and dorsal portions of the ring: forming the sides and roof of the neural canal.
  - iv. The spinous process or neurapophysis: a small blunt median process projecting backwards from the top of the neural arch.
  - v. The transverse processes: a pair of large lateral processes projecting from the sides of the neural arch.
  - vi. The articular processes or zygapophyses: on the anterior and posterior borders of the neural arch: articulate with corresponding processes on the vertebræ in front and behind, and so serve to link the vertebræ together.
    - a. The anterior articular processes or præzygapophyses: look upwards and slightly inwards.
    - β. The posterior articular processes or postzygapophyses: look downwards and slightly outwards.

### b. Peculiar vertebræ.

- i. The atlas or first vertebra: articulates in front with the posterior end of the skull: has no transverse processes. Note the large gap on the dorsal surface between the skull and the neural arch of the atlas: through this gap, which is closed by the strong occipito-atloid membrane, the central nervous system is divided and destroyed in the operation of pithing a frog.
- ii. The sacrum: the ninth vertebra: has very stoutbackwardly directed transverse processes, which support at their outer ends the pelvic arch.

c. The urostyle. The unsegmented posterior portion of the vertebral column, and probably equivalent to two or more vertebræ fused together. It articulates in front with the body of the sacral vertebra by two surfaces. Along its dorsal surface runs a prominent vertical ridge, largest in front and gradually diminishing posteriorly: the neural canal is continued down the anterior part of this ridge. At the sides of the urostyle, and about the length of a vertebra from its anterior end, are a pair of small holes through which nerves pass out, and which therefore correspond to inter-vertebral foramina.

II. The skull. Consists of (1) an axial portion, the cranium proper, enclosing the brain and forming an anterior continuation of the vertebral column: to this are fused (2) the olfactory capsules in front, and the auditory capsules behind:

(3) the bony framework of the jaws, and the hyoid apparatus.

In the skull the original cartilage has not been so completely replaced by bone as in the vertebral column, large tracts of unossified cartilage persisting in the adult. Besides the cartilage bones the skull is further strengthened by the addition of numerous membrane bones.

 The cranium proper. An unsegmented cartilaginous tube whose cavity forms the anterior part of the neural canal and lodges the brain. The roof of the tube is imperfect, there being one large anterior fontanelle, and two smaller posterior fontanelles, which are closed by membrane only. In the cartilage are developed cartilage bones, and around it membrane bones.

To study the cranium satisfactorily, the membrane bones should be stripped from one of the skulls you have prepared as directed above.

a. Cartilage bones of cranium proper.

i. The exoccipitals: two irregular bony masses at the sides of the posterior end of the skull. They bear the occipital condyles, two oval convex processes which articulate with the first vertebra or atlas; and almost completely surround the foramen magnum or entrance to the cranial cavity.

- ii. The sphenethmoid or girdle-bone: encircles the anterior end of the cranial cavity, and extends forwards into the olfactory region: in front it is divided by a vertical partition into right and left cavities, in which lie the olfactory nerves.
- b. Membrane bones of cranium proper.
  - i. The fronto-parietals: two long flat bones on the top of the brain-case; covering the fontanelles, and overlapping the hinder end of the sphenethmoid.
  - ii. The parasphenoid: a ⊥ shaped bone on the ventral surface of the cranium: its lateral processes underlying the auditory capsules.
- 2. The sense capsules: cartilaginous and bony capsules protecting the olfactory and auditory organs, and fused on to the sides of the cranium so as to form parts of the skull.
  - a. The auditory capsules are fused with the sides of the posterior end of the cranium, to which they form wing-like projections: they consist largely of cartilage.
    - a. Cartilage bones of auditory capsules.
      - The Pro-otics: a pair of irregular shaped bones in the anterior walls of the capsules, and forming also parts of their roof and floor.
  - b. The olfactory capsules are fused with the sides of the anterior end of the cranium, and also with one another. They consist very largely of cartilage, which is produced in front into the rhinal processes.
    - a. Cartilage bones of olfactory capsules. The sphenethmoid as already noticed extends forwards so as to invade the olfactory region, but does not properly belong to the olfactory capsules.
    - β. Membrane bones of the olfactory capsules.
      - i. The nasals: two triangular bones on the dorsal surface of the anterior end of the head: the bases of the triangles are turned towards the middle line, and meet one another in front. The posterior ends diverge from one another and enclose, with the anterior ends of the fronto-parietals, a diamond shaped patch in which the sphenethmoid is visible from the dorsal surface.

- ii. The vomers: two triradiate bones on the ventral surface of the fore part of the head: each vomer bears a small group of pointed teeth, and forms the inner boundary of the posterior narial opening of its side.
- 3. The jaws. Consist of two cartilaginous arches on each side, maxillary and mandibular, in connection with which cartilage and membrane bones are developed. Each arch meets and fuses with its fellow in the middle line in front; and the maxillary arches, forming the upper jaw, are firmly connected with the cranium by anterior and posterior bony struts.

## a. The maxillary arch.

- a. Cartilage bones of maxillary arch.
  - i. The pterygoid: a large triradiate bone, the inner limb of which is connected with the auditory capsule; while the posterior limb runs back to the angle of the mouth, and the anterior limb forwards along the upper jaw to
  - ii. The palatine bone: a slender transverse bone connecting the upper jaw with the anterior end of the sphenethmoid.
- iii. The quadratojugal: a short bone forming the posterior part of the margin of the upper jaw.

# B. Membrane bones of maxillary arch.

- i. The maxilla: a long thin bone forming the greater part of the margin of the upper jaw: bears teeth along its whole length which are anchylosed to the bone. It is connected behind with the quadratojugal; about the middle of its length with the anterior limb of the pterygoid and with the palatine; and in front with
- ii. The premaxilla: a small bone which meets its fellow in the middle line in front, and so completes the margin of the upper jaw: like the maxilla it bears teeth. It gives off on the dorsal surface a backwardly projecting process which forms part of the inner boundary of the nostril.

b. The mandibular arch: the upper part of the arch remains unossified as the quadrate cartilage, which forms the suspensorium, i.e., serves to connect the lower jaw with the skull. It is a rod of cartilage which is fused above with the auditory capsule, and runs downwards and backwards to the angle of the mouth, where it is connected with the quadratojugal bone. The lower part of the arch also persists in part unossified as Meckel's cartilage, which forms the base of the lower jaw, but is ensheathed by cartilage and membrane bones.

a. Cartilage bones of mandibular arch.

- i. The angulosplenial: ensheaths Meckel's cartilage at the sides and below along the greater part of its length: near its hinder end it is produced upwards into the coronary process.
- ii. The mentomeckelian: a small ossification in Meckel's cartilage at the symphysis, i.e., the union of the arches of the two sides at the chin.

B. Membrane bones of mandibular arch.

- i. The squamosal: a T shaped membrane bone, the body of which is closely applied to the outer surface of the quadrate cartilage. The posterior limb of the T is attached to the outer surface of the auditory capsule, and with the body of the squamosal helps to support the tympanic cartilage, a ring of cartilage surrounding the tympanic membrane.
- 4. The hyoid apparatus. Consists of the hyoidean arch and the remains of the branchial arches of the two sides, together with a median ventral plate—the body of the hyoid—which unites their lower ends together, and lies in the floor of the mouth. The whole apparatus consists almost entirely of cartilage.

a. The hyoid arch.

i. The columella: the top of the hyoid arch: forms a small rod, partly bone and partly cartilage, the inner end of which is inserted into the fenestra ovalis—an aperture in the outer wall of the auditory capsule—, while the outer end is attached to the tympanic membrane rather above its middle. ii. The anterior cornu of the hyoid: a long slender curved rod of cartilage: attached above to the auditory capsule just below the fenestra ovalis, and curving at first backwards and then forwards and downwards to be attached to the anterior outer angle of the body of the hyoid.

b. The body of the hyoid: a flat squarish plate of cartilage formed by the fused ventral ends of the hyoid and branchial arches, and lying in the floor of the mouth:

short processes are given off from its angles.

c. The posterior cornua of the hyoid: a pair of stout bony processes diverging from the hinder border of the body of the hyoid, and representing the lower ends of the fourth pair of branchial arches.

# B. The Appendicular Skeleton.

Consists of the limbs and the limb-girdles. As in the case of the axial skeleton it is at first entirely cartilage, which becomes afterwards replaced to a greater or less extent by cartilage-bone. Membrane bones, i.e., bones developed independently of cartilage, are very rare.

1. The pectoral girdle.

Consists originally of two half rings of cartilage, one on each side of the body, which they encircle a short way behind the head; the dorsal ends of the half rings are attached by ligaments and muscles to the vertebral column, while the ventral ends are united together in the median line by the sternum or "breast bone."

Each half ring bears in the middle of its outer surface a cupshaped cavity, which forms with the first bone of the fore-limb the shoulder joint. The part of the arch above the joint is the scapular portion; and the part below, which is divided into

anterior and posterior portions, the coracoid.

a. The scapular portion: divided into

i. The suprascapula: the upper portion: a thin expanded plate of cartilage, partly calcified and partly ossified, though very imperfectly.

ii. The scapula: an oblong bony plate, constricted in the middle: forms the upper half of the glenoid cavity, the cavity of the shoulder joint. b. The coracoid portion: divided into anterior and posterior portions, separated from one another by the coracoid foramen.

a. anterior part.

- The præcoracoid: a slender horizontal bar of cartilage connecting the anterior edge of the scapula with the sternum.
- ii. The clavicle: a slender membrane-bone, closely applied to the anterior border of the præcoracoid: its outer or scapular end is bent forwards almost at a right angle. Noteworthy as being the only membrane-bone in the appendicular skeleton.

B. posterior part.

- iii. The coracoid: a stout bone, wider at the inner than the outer end: connecting the posterior edge of the scapula with the sternum.
- c. The sternum: consists originally of two lateral halves which fuse completely in front and behind, but remain separate in the median portion. Presents from before backwards the following parts:
  - i. The episternum: a flat circular plate of cartilage.
  - ii. The omosternum: a slender bony rod projecting forwards in front of the clavicles.
  - iii. The epicoracoids: a pair of narrow strips of cartilage between the ventral ends of the præcoracoids and coracoids.
  - iv. The sternum proper: a rod of cartilage ensheathed in bone projecting backwards behind the epicoracoids.
  - The xiphisternum: a broad expanded plate of cartilage, bifid posteriorly.

#### 2. The fore-limb.

The bones, which are all cartilage bones, are elongated with enlarged ends, excepting the small bones of the wrist. The enlarged ends or epiphyses ossify independently of the shaft of the bone, with which they do not unite until late in life: the extremities are capped with articular cartilage. The end of a bone which, when the limb is extended, is nearer to the body is called its proximal end, the opposite extremity the distal end.

- a. The arm: in the arm there is only a single bone.
  - i. The humerus: the proximal end or head is enlarged, and articulates with the glenoid cavity of the pectoral girdle: below the head is the strong deltoid ridge extending along the proximal half of the anterior surface. At the distal end is a spheroidal articular surface for the bone of the fore-arm: and at either side of this a prominent condylar ridge, the inner or postaxial one being the larger of the two, especially in the male frog.

#### b. The fore-arm.

- i. The radio-ulna: corresponding to two bones, the radius and ulna in other animals: it is single at its proximal end, but in its distal half is imperfectly divided by a groove into anterior or radial, and posterior or ulnar portions. Its proximal end is hollowed out to articulate with the lower end of the humerus, and so form the elbow joint; beyond which it forms a backwardly projecting olecranon process.
- c. The wrist. Consists of six carpal bones arranged in two rows, proximal and distal, each row having three bones.
- d. The hand. Has four complete digits and a rudimentary pollex or thumb. Each digit consists of a proximal metacarpal bone, beyond which are a variable number of phalanges.
  - The pollex: consists simply of a small metacarpal bone.
  - ii. Corresponding to the fore-finger of man: consists of a metacarpal and two phalanges.
  - Corresponding to the middle-finger of man: consists of a metacarpal and two phalanges.
  - iv. Corresponding to the ring-finger of man: consists of a metacarpal and three phalanges.
  - Corresponding to the little-finger of man; consists of a metacarpal and three phalanges,

# 3. The pelvic girdle.

Consists primitively, like the pectoral girdle, of a couple of

half-rings of cartilage, fused together below and attached above to the tips of the transverse processes of the sacrum. In the adult frog the girdle is placed very obliquely so as to be nearly parallel with the vertebral column instead of at right angles to it.

Each half presents on its outer aspect a cup-shaped cavity the acetabulum—forming with the bone of the thigh, the hipjoint: we accordingly distinguish an iliac portion above the acetabulum, and an ischio-pubic portion below it, corresponding respectively to the scapular and coracoid divisions of the

pectoral girdle.

i. The ilium: forms the anterior and upper half of the acetabulum and extends forwards as an elongated rounded bar, which is attached in front to the transverse process of the sacrum, and bears along its dorsal surface a prominent vertical ridge of bone—the iliac crest, ending behind in an abrupt vertical border. Posteriorly the two ilia meet one another and are united together in the median line to form the iliac symphysis.

ii. The pubis: consists entirely of cartilage: forms the anterior portion of the ventral division of the girdle, and therefore corresponds to the præcoracoid in the pectoral girdle. The two pubes are completely fused together in the median plane, and form only a very small portion, about

one-sixth, of the acetabular cavities.

iii. The ischium: the posterior portion of the ventral division: corresponding therefore to the coracoid in the shoulder girdle. Forms the posterior third of the acetabulum. The two ischia are completely fused together in the median plane.

## 4. The hind-limb.

The bones have the same general characters as those of the fore-limb, to which they correspond very closely.

a. The thigh.

i. The femur: a long slender bone expanded at both ends, and curved in a somewhat sigmoid manner. The proximal end or head is spheroidal, and fits into the acetabulum to form the hip joint: the distal end is somewhat expanded laterally.

## b. The leg.

- i. The os cruris or tibio-fibula: a single bone rather longer than the femur, slightly curved, and expanded laterally at both ends. Presents along the greater part of its length a distinct groove indicating its correspondence with two bones, tibia and fibula, which in many animals remain distinct from one another.
- c. The ankle. Corresponding to the wrist in the fore-limb: consists like this latter of two rows of tarsal bones.
  - a. Proximal row of tarsal bones: consists of two elongated bones united together at both ends, but widely separated in the middle.
    - i. The astragalus: on the præaxial or tibial side.
    - ii. The calcaneum: on the postaxial or fibular side.
  - β. Distal row of tarsal bones: consists of two very small bones.
- d. The foot. Has five complete digits, and a supernumerary toe as well. Each digit consists of a proximal metatarsal bone, beyond which are a variable number of phalanges.
  - i. The hallux, corresponding to the great toe of man: the præaxial digit, and the smallest of the series. Consists of a metatarsal and two phalanges.

On the inner side of the hallux is the calcar, supposed to be an additional or supernumerary

- Consists of a metatarsal and two phalanges.
   Consists of a metatarsal and three phalanges.
- iv. The longest of the toes: consists of a metatarsal and four phalanges.

v. The postaxial digit, corresponding to the little toe in man: consists of a metatarsal and three phalanges.

# Chap. V .- THE MUSCULAR SYSTEM OF THE FROG.

The muscles, or flesh, are the direct means by which the various movements of the body and its several parts are brought about. A muscle usually consists of a fleshy belly which is attached at each end by means of tendons to some hard part, very commonly to bone. Motion is effected by the muscle contracting, i.e., shortening, and so bringing its two ends, and consequently the parts to which the ends are attached, nearer together. Of the two attachments of a muscle one is usually to a more fixed and central part, the other to a more movable and peripheral part: the former attachment is called the origin of the muscle, the latter its insertion.

Muscles are of two kinds: (1) voluntary muscles, i.e., those over which we have direct control, as the muscles of the arm; and (2) involuntary muscles, i.e., those over which we have no direct control, as the muscles of the heart and bloodvessels, or of the alimentary canal.

Voluntary muscles, which are the only ones dealt with in this chapter, are usually attached at both ends to bone; but one or other end, or both, may be attached to aponeuroses, strong connective tissue membranes, ensheathing the muscles and other parts, or separating them from one another.

In dissecting the muscles, take a frog that has been in spirit for a day or more. When cleaning a muscle be careful to put it on the stretch, and to dissect along and not across its fibres: define the origin and insertion of the muscle very clearly, and test its action by pulling it gently with the forceps in the direction of its fibres. Always have the skeleton in front of you so as to see accurately the origins and insertions of the muscles.

#### A. Muscles of Trunk.

# 1. Muscles of ventral body-wall.

Pin out the frog on its back, remove the skin, and clean the ruscles.

i. The rectus abdominis: runs longitudinally along the midventral wall, the muscles of the two sides being separated in the middle line by the linea alba, a longitudinal band of connective tissue, immediately dorsad of which lies the anterior abdominal vein. Each rectus muscle is divided into bellies by five transverse tendinous intersections.

The muscle arises from the pubis, runs forward and is inserted into the dorsal surface of the sternum and coracoid.

- ii. The pectoralis: a large fan-shaped muscle consisting of a thoracic portion, which arises from the whole length of the ventral surface of the sternum; and an abdominal portion arising from the aponeurosis along the outer side of the rectus abdominis almost as far back as the pubis. From this extensive origin the fibres converge to the deltoid ridge of the humerus into which they are inserted, the line of insertion extending down almost to the elbow.
- iii. The obliquus externus: a thin sheet of muscle which arises from the aponeurosis of the back, a short distance on either side of the vertebral column, and covers the whole of the side of the body, the fibres running obliquely downwards and backwards to end in an aponeurosis which passes dorsad of the rectus abdominis to be inserted into the linea alba.
- iv. The obliquus internus: lies beneath the obliquus externus, which must be removed in order to see it. Arises from the transverse processes of the vertebræ from the fourth backwards, and from the ilium. The fibres run downwards and forwards, and are inserted in front into the coracoid and sternum, some of the fibres passing round the esophagus and pericardium. The hinder two-thirds of the muscle pass dorsad of the tendon of the obliquus externus and are inserted like it into the linea alba.

## 2. Muscles of Back.

Pin out the frog on its belly: remove the skin, and clean the muscles in order.

- i. The depressor mandibuli: a broad triangular muscle which arises from the fascia dorsalis lying dorsal of the scapula: the fibres run downwards behind the tympanic membrane, and converge to be inserted into the angle of the lower jaw. The muscle by its contraction opens the mouth.
- ii. The cucultaris: a small muscular band which, arising from the exoccipital near the middle line, runs backwards and outwards, and is inserted into the dorsal border of the suprascapula.
- iii. The latissimus dorsi: a triangular muscle behind the depressor mandibuli: it arises from the fascia dorsalis just behind the shoulder girdle, its origin being partly covered by the obliquus externus. The fibres run outwards and forwards, converging to be inserted by a long tendon into the deltoid ridge of the humerus.

Dissect away the depressor mandibuli and latissimus dorsi from their origins, and turn them down.

iv. The infraspinatus arises from the dorsal surface of the suprascapula, partly overlapped by the latissimus dorsi; it runs outwards to be inserted into the deltoid ridge of the humerus; its action is to elevate the arm.

Lift up the suprascapula and note the muscles attaching it to the body, viz.:

v. The retrahens scapulæ, behind.

vi. The levator anguli scapulæ, in front.

Remove the suprascapula on one side, and clean the median longitudinal muscles of the back.

vii. The extensor dorsi communis: a longitudinal mass of muscle arising from the urostyle, and running forwards and slightly outwards: inserted into the ilium, into the transverse processes of the vertebræ, and in front into the posterior end of the skull. The anterior part is divided by transverse tendinous intersections.

- viii. The intertransversales: small muscles running between the transverse processes of the vertebræ and lying beneath the extensor communis.
- ix. The glutæus: arises from the outer side of the posterior two-thirds of the ilium, and runs backwards to be inserted into the trochanter of the femur.

#### B. Muscles of Head.

## 1. Muscles of the ventral surface of the head.

i. The mylohyoid or submandibular muscle: a flat sheet of muscle running across from one ramus of the mandible to the other, and divided down the middle line by a tendinous intersection. A narrow strip along the posterior border is commonly separated by a slight interval from the major or anterior part of the muscle.

Remove the mylohyoid muscle, and the sternal portion of the

pectoralis.

- ii. The geniohyoid: a narrow longitudinal band a short distance from the middle line: arises from the lower jaw close to the chin, runs back on the ventral surface of the body of the hyoid, and divides posteriorly into two portions which are inserted respectively into the bony and the cartilaginous posterior processes of the hyoid.
- iii. The sternohyoid: is practically the anterior continuation of the rectus abdominis. Arises from the dorsal surface of the coracoid and clavicle, and is inserted into the ventral surface of the body of the hyoid, the tendon passing between the two divisions of the geniohyoid.
- iv. The hyoglossus: arises on either side from the posterior bony horn of the hyoid: the two muscles converge and meet in front of the larynx, bounding with the diverging aortic arches behind a lozenge-shaped patch in which is the larynx. In front of the larynx the muscle runs forward in the middle line as a stout band nearly to the chin: it then enters the tongue, and runs back along it to its tip.

v. The petrohyoid: a set of four muscular bands which arise close together from the outer surface of the auditory capsule, and diverging from one another in a fan-like manner, pass round the floor of the pharynx and esophagus to be inserted in front into the median ventral line of the pharynx, and behind into the side of the hyoid. The first or most anterior band is a wide thin sheet of muscular tissue, while the three posterior portions are very narrow slips.

#### 2. Muscles of the side of the head.

Remove the skin carefully from the side of the head and jaws, noticing how much more closely it is attached to the underlying parts than was the case in the body.

- a. Depressors of the lower jaw: opening the mouth.
  - The depressor mandibuli: has been already seen and dissected.
- b. Elevators of the lower jaw: shutting the mouth: lie in the space between the auditory capsule and the eye.
  - ii. The temporalis: arises from the upper surface of the auditory capsule; passes outwards and downwards between the pterygoid and maxillary bones and in front of the cartilaginous ring supporting the tympanic membrane (from which some of its fibres arise): is inserted into the coronoid process on the inner side of the lower jaw.
  - iii. The pterygoideus: a slender muscle placed just in front of the temporalis and partly covered by it: arises from the side wall of the skull, and is inserted into the mandible further back than the temporalis and very close to the joint.
  - iii. The masseter: a small short muscle placed behind the temporalis: it arises from the quadratojugal and runs downwards and slightly backwards to be inserted into the outer surface of the mandible, just in front of the joint.

To see the insertions of these last three muscles the mouth should be opened widely.

## 3. Muscles of the eyeball.

Remove the temporal and pterygoid muscles carefully, dissecting them away from their origins, and then turning the muscles down and cutting them short close to their insertions. Remove also the lower jaw: pin the frog out on its back and dissect away carefully the mucous membrane of the roof of the mouth: note

i. The levator bulbi: a thin sheet of muscle lying between the mucous membrane and the eye. Its fibres arise from the side of the skull, run outwards underneath the eye and are inserted into the upper jaw. The muscle by its contraction serves to lift up the eyeball and so make it more prominent. Some of its fibres are inserted into the lower eyelid which they serve to depress, acting as a depressor palpebræ inferioris.

Remove the levator bulbi and clean the remaining muscles, dissecting them partly from the dorsal and partly from the ventral side.

- a. The recti muscles: a group of four small muscles which arise close together from the inner and posterior angle of the orbit close to the optic foramen, and run forwards and outwards, diverging from one another, to be inserted into the bulb of the eye.
  - i. The rectus superior: inserted into the dorsal surface of the eyeball: seen best from above.
  - ii. The rectus externus: the most posterior of the four: inserted into the posterior surface of the eyeball: seen best from the side or from below.
  - iii. The rectus internus: the longest of the four: runs forward between the skull wall and the eyeball, and is inserted into the inner or median side of the eye: seen best from below.
  - iv. The rectus inferior: inserted into the under surface of the eyeball: seen best from below.
- b. The obliqui muscles: a set of two small muscles which arise close together from the palatine bone at the anterior end of the orbit, and run backwards to be inserted into the eyeball, one above and one below.

- i. The obliquus superior: inserted into the dorsal surface of the eyeball just in front of the rectus superior: seen best from above.
- ii. The obliquus inferior: passes backwards beneath the rectus internus, and is inserted into the eyeball between it and the rectus inferior: seen best from below.
- c. The retractor bulbi, or choanoid muscle: a funnel-shaped muscle which lies within the four recti and embraces the optic nerve: it arises from the parasphenoid, and is inserted into the eyeball. Best seen from below by carefully removing the recti muscles.

#### C. Muscles of Hind-limb.

If the frog's leg be stretched back as in the act of swimming, we distinguish dorsal and ventral surfaces, an outer border in which is the projection of the knee, and an inner border along which is the bend of the knee. The outer border, which corresponds to the front of the leg in man, is called the extensor surface, inasmuch as the muscles which extend or straighten the leg lie along this edge: the inner border is the flexor surface. The "ventral" and "dorsal" surfaces only appear to be such in consequence of the extreme obliquity of the pelvic girdle: they are really anterior and posterior, and are better called præaxial and postaxial: they correspond respectively with the inner and outer surfaces of the human leg. If the foot be examined carefully, it will be seen that the first digit or "big toe" is on the præaxial side, and hence may be called the præaxial digit: while the fifth or "little toe" is on the postaxial side, and is therefore the postaxial digit.

## 1. Muscles of thigh.

Remove the skin from one of the legs of the frog and clean the muscles first of the præaxial and then of the postaxial surfaces.

- a. Superficial muscles of præaxial (apparent ventral) surface.
  - i. The sartorius: a long narrow muscular band which crosses the thigh somewhat obliquely. Arises from the iliac symphysis below the acetabulum, and is inserted into the inner side of the head of the tibia.

- ii. The adductor magnus: a large muscle lying along the inner border of the sartorius, but passing beneath it at its lower end. Arises from the pubic and ischial symphyses, and passes under the sartorius to be inserted into the lower third of the femur.
- iii. The adductor longus: a long narrow muscle lying along the outer side of the adductor magnus, and almost completely hidden by the sartorius: arises from the iliac symphysis beneath the sartorius, and unites a little below the middle of the thigh with the adductor magnus.
- iv. The rectus internus major: a large muscle lying along the inner side of the adductor magnus and the sartorius. Arises from the ischial symphysis and is inserted into the head of the tibia.
- v. The rectus internus minor; a narrow flat band of muscle running along the inner (flexor) margin of the thigh: arises from a tendinous expansion connected with the ischial symphysis, and is inserted into the inner side of the tibia just below its head.

# b. Superficial muscles of extensor surface of thigh.

- i. The triceps extensor femoris: the great extensor muscle of the thigh: arises by three distinct heads, which may be described separately, and is inserted into the tibia just below its head.
  - a. The rectus anticus femoris: forms the middle head of the triceps: arises from the ventral border of the posterior third of the ilium, in front of the acetabulum: about half way down the thigh it joins
  - β. The vastus internus: the præaxial division of the triceps: a large muscle arising from the ventral and anterior border of the acetabulum, and lying in the thigh between the sartorius and the rectus anticus.

- γ. The vastus externus: the postaxial division of the triceps: arises from the posterior edge of the dorsal crest of the ilium: joins the other two divisions of the triceps about the junction of the middle and lower thirds of the thigh.
- Superficial muscles of the postaxial (apparent dorsal) surface of the thigh.
  - The glutæus: has been already noticed: lies in the thigh between the rectus anticus and vastus externus.
  - ii. The biceps: a long slender muscle which arises from
    the crest of the ilium just above the acetabulum:
    it lies in the thigh along the inner border of the
    vastus externus, and is inserted by a flattened
    tendinous expansion into the lower end of the
    femur and the head of the tibia.
  - iii. The semimembranosus: a stout muscle lying along the inner side of the biceps, between it and the rectus internus minor. Arises from the dorsal angle of the ischial symphysis just beneath the cloacal opening, and is inserted into the back of the head of the tibia. It is divided about its middle by an oblique tendinous intersection.
  - iv. The pyriformis: a slender muscle which arises from the tip of the urostyle, passes backwards and outwards between the biceps and the semimembranosus, and is inserted into the femur at the junction of its upper and middle thirds.

# d. Deep muscles of the thigh.

Lay the frog on its back so as to dissect the thigh from its præaxial side. Separate the adductor magnus and rectus internus major with blunt instruments so as to expose:—

i. The semitendinosus: a long thin muscle which arises by two heads; an anterior one from the ischium close to the ventral angle of the ischial symphysis and the acetabulum; and a posterior one from the ischial symphysis. The anterior head passes through a slit in the adductor magnus and unites with the posterior head in the lower third of the thigh. The tendon of insertion is long and thin, and joins that of the rectus internus minor to be inserted into the tibia just below its head.

Divide the adductor magnus and the sartorius in the middle, and turn the cut ends backwards and forwards, so as to expose

- ii. The adductor brevis: a short wide muscle lying beneath the upper end of the adductor magnus. It arises from the pubic and ischial symphyses, and is inserted into the præaxial surface of the upper half of the femur.
- iii. The pectineus: a rather smaller muscle, lying along the outer (extensor) side of the adductor brevis. It arises from the anterior half of the pubic symphysis in front of the adductor brevis, and is inserted like it into the upper half of the femur.
- iv. The ileo-psoas: arises by a wide origin from the inner surface of the acetabular portion of the ilium: it turns round the anterior border of the ilium, and crosses in front of the hip joint, where for a short part of its course it is superficial between the heads of the vastus internus and rectus anticus femoris: it then passes down the thigh beneath these muscles, and is inserted into the back of the upper half of the femur.
- v. The quadratus femoris: a small muscle on the back of the upper part of the thigh: arises from the ilium above the acetabulum, and from the base of the iliac crest: it lies beneath the pyriformis and behind the biceps, and is inserted into the inner surface of the upper third of the femur between the pyriformis and the ileo-psoas.
- vi. The obturator: a deeply situated muscle which arises from the whole length of the ischial symphysis and the adjacent parts of the iliac and pubic symphyses, and is inserted into the head of the femur close to the glutæus.

2. Muscles of the leg.

As in the thigh we distinguish extensor and flexor surfaces, corresponding to the front and back of the leg in man; and also preaxial and postaxial surfaces, corresponding to the inner and outer sides of the human leg.

Lay the frog on its belly and commence the dissection from the

postaxial surface.

- i. The gastrocnemius: the large muscle forming the calf of the leg: has two heads of origin, whereof the larger one arises by a strong flattened tendon from the flexor surface of the lower end of the femur; while the smaller head, which joins the main muscle about one-fourth of its length below the knee, arises from the edge of the tendon of the triceps extensor femoris where it covers the knee. The muscle is thickest in its upper third, and tapering posteriorly ends in the strong tendo Achillis, which passes under the ankle joint, being much thickened as it does so, and ends in the strong plantar fascia of the foot.
- ii. The tibialis posticus: arises from the whole length of the flexor surface of the tibia: ends in a tendon which passes round the internal malleolus, lying in a groove in the lower end of the tibia, and is inserted into the dorsal surface of the astragalus.
- iii. The tibialis anticus: lies on the extensor surface of the leg: arises by a long thin tendon from the lower end of the femur, and divides about the middle of the leg into two bellies which are inserted into the proximal ends of the astragalus and calcaneum respectively.
- iv. The extensor cruris brevis: lies along the præaxial side of the tibialis anticus, partly covered by it and partly by the strong fascia of the leg. Arises by a long tendon from the præaxial condyle of the femur, runs in a groove in the upper end of the tibia, and is inserted into the extensor surface of the tibia along nearly its whole length.

v. The peroneus: a stout muscle which lies along the postaxial surface of the leg, between the tibialis anticus and the gastrocnemius. It arises from the lower end of the femur, and is inserted into the external malleolus of the tibia and the proximal end of the calcaneum.

## Chap. VI .- THE NERVOUS SYSTEM OF THE FROG.

The nervous system consists of (1) a central portion—the brain and spinal cord—which lies in the cartilaginous and bony tube formed by the cranium and vertebral column, and which is the centre where sensations are felt, and whence motor impulses causing the muscles to contract take their origin: and (2) a peripheral portion—the nerves themselves—which connect the central portion with the skin, sense organs, muscles, viscera, etc., and so serve to convey sensory impulses from these parts to the brain and cord, or motor impulses from the central organs to the muscles.

These two functions are fulfilled by different nerves, which may accordingly be distinguished as (a) afferent or sensory, nerves, conveying impulses to the central parts, and (b) efferent or motor nerves, conveying impulses from the brain or cord.

A special set of nerves in connection with the bloodvessels

and viscera form the sympathetic nervous system.

For the dissection of the nervous system specimens should be chosen which have been in strong spirit for two or three days, and in which the brain has been exposed to the action of the spirit by removal of the roof of the skull.

## A. The Central Nervous System.

Divisible into an anterior portion—the brain—lying in the cavity of the cranium; and a posterior portion—the spinal cord—which lies in the neural canal of the vertebral column. There is no sharp line of demarcation between the two portions, which

are directly continuous with one another.

If the brain and spinal cord have not been already exposed, clear away the dorsal muscles from both sides of the spine: cut through the occipito-atloid membrane, flexing the frog's head slightly to make the membrane tense, and being careful not to injure the brain beneath it. Introduce one blade of the scissors into the cranial cavity, with the flat surface of the blade parallel to the back of the frog, and keeping as close to the roof of the skull as possible. Cut carefully through the side walls of the skull, first on one side and then on the other. Turn the roof of the skull forwards with forceps, and remove it altogether.

Similarly cut through and remove the neural arches of the vertebræ one by one from before backwards.

Examine and draw the central nervous system in situ, showing

#### 1. The brain.

- a. The dorsal surface of the brain: note from before backwards the following parts, removing the pigmented membrane—pia mater—covering the several parts as you come to them.
  - i. The olfactory lobes: united together in the middle line by a median commissure: give off the olfactory nerves from their anterior ends: are separated behind by slight constrictions from
  - ii. The cerebral hemispheres: smooth ovoid bodies which touch one another in the median line but are not fused together. Behind the hemispheres and between their diverging ends is
  - iii. The thalamencephalon: lozenge-shaped: covered by a thick vascular membrane—the choroid plexus—through which passes the stalk of the pineal gland, a small body adherent to and removed with the roof of the skull. On removing the choroid plexus a small hole is seen in the roof of the thalamencephalon, connecting the hollow stalk of the pineal gland with the third ventricle. The thickened sides of the thalamencephalon are the optic thalami.
  - iv. The optic lobes: two prominent ovoid bodies touching one another in the median line: form the widest part of the brain.
  - v. The cerebellum: a narrow transverse band immediately behind the optic lobes.
  - vi. The medulla oblongata: the part of the brain behind the cerebellum: is widest in front and gradually tapers towards its posterior end, where it is continuous with the spinal cord. It is covered by a very vascular triangular membrane, on removing which the fourth ventricle is exposed.

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- iv. The crura cerebri: two dense white columns of nervous matter, lying at the base of the optic lobes, and partly hidden by the pituitary body: serve to connect the hemispheres with the medulla and spinal cord,
- v. The anterior fissure of the brain: a median longitudinal groove on the under surface of the hinder part of the brain: continuous with a similar groove on the under surface of the spinal cord.

#### 2. The spinal cord.

A somewhat flattened band presenting brachial and lumbar enlargements opposite the points of origin of the nerves for the fore and hind limbs respectively, and slightly constricted between these two points. About the level of the sixth vertebra the cord narrows rapidly to form a fine thread, the filum terminale, which extends back into the canal of the urostyle. Besides the anterior or ventral fissure the cord presents a dorsal fissure, a median longitudinal groove starting from the hinder border of the fourth ventricle and extending to the commencement of the filum terminale.

## B. The Peripheral Nervous System.

I. The Spinal Nerves: Ten pairs of nerves arise from the sides of the spinal cord; each nerve arising by two roots, ventral or "anterior," and dorsal or "posterior," which unite together at their points of exit from the vertebral canal through the intervertebral foramina: just before their union the pos-

terior root bears a ganglionic swelling.

Within the vertebral canal the roots of the anterior spinal nerves run nearly transversely outwards, so as to leave the canal opposite their points of origin from the spinal cord. The roots of the middle and posterior nerves, owing to the vertebral column being of greater length than the part of the cord belonging to it, pass obliquely backwards to their points of exit: and in the case of the hindmost nerves, the roots run backwards within the vertebral canal some distance before reaching their foramina of exit, forming with the filum terminale the cauda equina.

Slice off the upper surface of the brain, to expose

#### b. The cavities of the brain.

- The lateral ventricles: extend through the whole length of the cerebral hemispheres and a short way into the olfactory lobes.
- ii. The third ventricle: situated in the thalamencephalon: opens in front through the foramina of Munro into the lateral ventricles: opens above into the stalk of the pineal gland; and presents in the hinder part of its floor a depression, the infundibulum.
- iii. The aquæductus sylvii or iter a tertio ad quartum ventriculum: a narrow passage leading from the third to the fourth ventricles: communicates above with the cavities or ventricles of the optic lobes, which are hollow.
- iv. The fourth ventricle: the large triangular cavity in the medulla already exposed by removal of the vascular membrane covering it.

#### c. The ventral surface of the brain.

Cut through the medulla at the level of the hinder end of the skull: carefully remove the brain from the cranial cavity, noting the several nerves arising from it, and cutting through these as close to the skull wall as possible. Lay the brain on its dorsal surface, and examine and draw the ventral surface, showing:—

 The optic chiasma: formed by the roots of the optic nerves crossing one another; the point of crossing being opposite the hinder ends of the hemispheres.

Trace back the optic nerves behind their point of crossing to their origins from the optic lobes.

- ii. The tuber cinereum: a small median swelling immediately behind the optic chiasma, and caused by the depression of the floor of the third ventricle to form the infundibulum.
- iii. The pituitary body: a flattened sac, placed behind and continuous with the tuber cinereum. Is very liable to be left behind in the skull on removing the brain.

4, 5, and 6: the fourth, fifth, and sixth spinal nerves are small, and supply the muscles and skin of the body-wall. They leave the vertebral canal between the fourth and fifth, fifth and sixth, and sixth and seventh vertebræ

respectively.

7, 8, and 9: the seventh, eighth, and ninth spinal nerves together form the sciatic plexus. The roots of these three nerves within the vertebral canal form the main part of the cauda equina. The seventh nerve leaves the canal between the seventh and eighth vertebræ, the eighth nerve between the eighth and ninth vertebræ, and the ninth nerve between the ninth or sacral vertebra and the urostyle. Outside the vertebral canal the three nerves unite together opposite the middle of the urostyle to form the sciatic plexus, from which branches are given to the large intestine, bladder, oviducts, etc. Just before joining the plexus the seventh nerve gives off the ileohypogastric and crural nerves supplying the muscles and skin of the abdomen and thigh. Beyond the plexus is the large sciatic nerve, which runs down the thigh, giving branches to it, and divides shortly above the knee into the tibial and peroneal nerves supplying the leg and foot.

10: The coccygeal or tenth spinal nerve emerges through a small hole in the side of the urostyle near its anterior end. It gives branches to the bladder, cloaca, and other adjacent parts.

II. The Sympathetic Nervous System. Consists of a longitudinal nervous band on either side of the body connected by branches with the several spinal nerves. The two main sympathetic trunks lie at first close to the dorsal surface and alongside the vertebral column: further back they leave the vertebral column and run alongside the dorsal aorta.

Each trunk receives a branch from each of the spinal nerves of its side, and at the junction of each of these branches with

the main trunk there is a ganglionic enlargement,

- a. The spinal nerves within the vertebral canal: note
  - i. The roots of the nerves: dorsal and ventral.
  - ii. The obliquity of the middle and posterior roots.
  - The cauda equina: formed by the roots of the hinder nerves together with the filum terminale.
  - iv. The ganglia on the posterior roots as they pass through the intervertebral foramina: best seen from the ventral surface, where they are covered over by whitish calcareous patches, which form conspicuous objects on either side of the vertebral column. Remove the patches carefully to see the ganglia.

#### b. The spinal nerves outside the vertebral canal:

Lay the frog on its back: cut through and pin out the body-walls, and remove the abdominal viscera. Note the spinal nerves, seen as white cords at the sides of the vertebral column. Clean the nerves on one side and follow them up to their distribution. Each nerve divides directly after the union of its two roots into a small dorsal branch and a much larger ventral branch.

- The hypoglossal or first spinal nerve: leaves
  the vertebral canal between the first and
  second vertebræ, and then runs forward on the
  under surface of the head beneath the mylohyoid and in the substance of the geniohyoid
  muscle to the chin, where it enters the tongue
  in which it ends. Supplies the muscles of the
  tongue and floor of the mouth, and also some
  of the muscles of the back and shoulder.
- 2 and 3: the second and third spinal nerves, leaving the canal between the second and third, and third and fourth vertebræ respectively; unite together to form the brachial nerve, which gives off a large coracoclavicular branch to the shoulder muscles and then runs down the arm, supplying it with muscular and cutaneous branches, and divides just above the elbow into the radial and ulnar nerves which supply the forearm and hand.

3. The motor oculi: a small nerve arising from the ventral surface of the brain close to the median line and between the crura cerebri. It supplies four of the muscles moving the eyeball; viz., the rectus superior, rectus internus, rectus inferior, and obliquus inferior.

Owing to its small size the third nerve is not easy to dissect in the frog.

- 4. The pathetic or trochlear nerve: a very slender nerve, arising from the dorsal surface of the brain between the optic lobes and the cerebellum, and supplying the obliquus superior muscle of the eyeball.
- 5. The trigeminal: the largest of the cranial nerves in the frog. Arises from the side of the anterior part of the medulla, and runs outwards and forwards to the skull wall; just before reaching this it expands into a large swelling—the Gasserian ganglion. It then passes through the skull wall immediately in front of the auditory capsule, and divides at once into two main branches:
  - i. The ramus ophthalmicus: runs forward through the orbit lying close to its inner side, between the skull wall and the eye. It lies beneath the rectus superior, but above all the other muscles of the eyeball and the optic nerve. At the anterior end of the orbit it divides into two branches which pass through the walls of the nasal capsule, and supply the skin of the fore part of the head.

To trace this branch dissect from the dorsal surface: cut away carefully with scissors the side wall of the cranium; cut through and turn aside the rectus superior, and find the nerve running close alongside the skull wall, between it and the eyeball. Trace it forwards to the nose.

ii. The ramus maxillo-mandibularis: runs directly outwards behind the eyeball, in front of the auditory capsule and between the temporal and pterygoid muscles. After a very short course it divides into the maxillary and mandibular nerves. The coccygeal or tenth spinal nerve, unlike the others, is connected with the sympathetic by more than a single branch: the actual number of these branches is not constant, but is said to vary from two up to as many as twelve.

From the sympathetic ganglia nerves are given off to the bloodyessels and viscera, the chief of which are

- i. The cardiac plexus: formed by nerves arising from the first sympathetic ganglion: the plexus is a meshwork of nerves on the auricles and around the great vessels at their openings into the heart.
- ii. The solar plexus: on the dorsal surface of the stomach: the nerves are derived mainly from the third, fourth, and fifth ganglia. Hepatic, renal, genital, hæmorrhoidal, and vesical plexus also exist in connection with the liver, kidney, reproductive organs, large intestine, and bladder respectively.

III. The Cranial Nerves. There are ten pairs of cranial nerves in the frog, which are numbered in order from before backwards.

To dissect the cranial nerves expose the brain by removing the roof of the skull as already described, and then follow the special instructions given in the case of the more important nerves.

1. The olfactory nerves: the special nerves of smell: arise from the anterior ends and sides of the olfactory lobes, and are distributed to the membrane lining the nasal cavities.

To see their course and distribution dissect from the dorsal side, removing the roof of the anterior part of the skull, including the nasal bones.

2. The optic nerves: the nerves of sight: arise from the sides of the brain just below the optic lobes, cross one another partially at the optic chiasma on the under surface of the brain, and then run outwards to the eyeballs.

The course of the optic nerves has been fully seen in previous dissections.

i. The ramus palatinus; runs forward in the floor of the orbit a short distance from the side wall of the skull, and immediately beneath the mucous membrane of the floor of the mouth. Near the anterior end of the orbit it divides into two branches, one of which runs outwards and anastomoses with the ramus maxillaris of the trigeminal nerve, while the other runs forwards to the anterior part of the roof of the mouth. Supplies the mucous membrane of the roof of the mouth.

Dissect this nerve from the ventral surface: cut away the lower jaw: carefully remove the mucous membrane of the roof of the mouth, and find the nerve lying on the ventral surface of the eyeball and its muscles, and running parallel to and a short distance from the skull wall. Trace it backwards and forwards.

ii. The ramus hyomandibularis: runs outwards and backwards round the front end of the auditory capsule; it then crosses over the inner end of the columella and turns downwards in the posterior wall of the Eustachian tube to the angle of the mouth, giving branches to the tympanic membrane and to the articulation of the mandible. It then divides into two branches.

The dissection of this nerve, which is not easy, may be performed thus:—remove the shoulder-girdle of one side; also the depressor mandibuli and temporal muscles: open the cranial cavity as before, to expose the brain; remove the tympanic membrane and clean the outer end of the columella. Cut away carefully the roof of the auditory capsule by a horizontal cut, just above the level of the columella: find the facial nerve running round the front end of the auditory capsule and in close contact with it, and trace it back over the columella and down to the angle of the mouth.

a. The ramus mandibularis: runs forward in the floor of the mouth, lying along the inner edge of the lower jaw and between the mylohyoid muscle and the skin, as far forward as the chin Dissect from the ventral surface; remove the skin from the under surface of the floor of the mouth and find the nerve running along the inner border of the mandible.

- β. The ramus hyoideus: the posterior and larger of the two branches: runs forward in the floor of the mouth along the anterior cornu of the hyoid, supplying its muscles.
- 8. The auditory nerve: the nerve of hearing: arises from the side of the medulla immediately behind and in close contact with the root of the facial nerve: enters the auditory capsule and ends in the internal ear.
- 9. The glossopharyngeal nerve: arises from the side of the medulla behind the auditory nerve, by a root common to it and the tenth nerve: leaves the skull by an aperture immediately behind the auditory capsule, and divides behind the capsule into two branches:
  - i. The ramus anterior: runs downwards and forwards round the auditory capsule and beneath the depressor mandibuli muscle to join the facial nerve just after it has crossed over the columella.

The dissection already made for the ramus hyomandibularis of the facial nerve will show also the above branch of the glossopharyngeal.

> ii. The ramus posterior: runs downwards and forwards to the ventral wall of the pharynx, passing beneath the fourth division of the petrohyoid muscle but superficial to the others; it runs just behind and parallel to the anterior cornu of the hyoid. On reaching the floor of the mouth it crosses obliquely dorsad of the hypoglossal or first spinal nerve, and then runs forwards in a peculiarly sinuous course, close to the middle line and between the geniohyoid and hyoglossus muscles, to the base of the tongue which it enters and in It supplies the petrohyoid which it ends. muscle, and the mucous membrane of the pharynx and tongue.

The dissection of the first part of the nerve is best performed from the side, and is much facilitated by distending the esophagus and pharynx with a cork or roll of paper. Its course along the floor of the mouth to the tongue should be dissected from the ventral side.

- 10. The pneumogastric or vagus nerve: arises, as already noticed, in common with the glossopharyngeal. It leaves the skull by the same aperture as the ninth nerve, and immediately outside the skull presents a ganglionic enlargement: it gives off dorsal branches to the muscles of the back, and then runs backwards and downwards round the side wall of the pharynx, running along the hinder border of the fourth division of the petrohyoid muscle: behind this muscle it divides into its main branches.
  - i. The ramus laryngeus: loops round the posterior cornu of the hyoid and round the pulmocutaneous artery close to its origin from the aortic trunk: passes inwards dorsad of the artery to the middle line where it ends in the larynx.
  - ii. The ramus cardiacus: passes dorsad of the pulmonary artery to the interauricular septum of the heart.
  - iii. The rami pulmonales: follow the course of the pulmonary artery to the lung, in which they end.
  - iv. The rami gastriei: usually two in number: run through the partial diaphragm formed by the anterior fibres of the obliquus internus muscle, and end in the walls of the stomach.

The dorsal portions of the several branches of the vagus are best dissected from the side: to see them properly, the shoulder girdle and fore-limb must be removed and the esophagus well distended: the terminal branches must be dissected from the ventral surface.

# IV. The Cranial Portion of the Sympathetic Nervous System.

The main sympathetic trunk of each side extends forwards in front of the first ganglion, and enters the skull at the foramen in the exoccipital bone through which the glossopharyngeal and vagus nerves pass out: it is connected with the vagus nerve, and then runs forwards within the skull to the Gasserian ganglion of the trigeminal nerve in which it ends.

## C. Histology of Merves.

Nervous matter consists histologically of elements of two kinds, nerve cells and nerve fibres. The nerve cells are branching nucleated cells connected by their processes with one another and with the nerve fibres. The nerve cells are the centres whence impulses originate, while the nerve fibres serve to convey those impulses from one part to another. A local accumulation of nerve cells is called a ganglion.

- Merve Pibres: are of two kinds, medullated and nonmedullated.
  - a. Medullated nerve fibres: form the greater part of the cranial and spinal nerves.

Take a small piece of the sciatic or some other nerve from a freshly killed frog: spread out and tease in a drop of normal salt solution: examine with low and high powers: note

- i. The nerve fibres: unbranched.
- ii. The **perineurium**, or connective tissue binding the nerve fibres into bundles or "nerves."

In each nerve fibre note

- iii. The primitive sheath, or sheath of Schwann: a very delicate external investment, seen with difficulty, and only in certain places.
- iv. The medullary sheath: a thick fatty layer within the primitive sheath.

Tease in glycerine a small piece of nerve that has been treated with osmic acid: examine with high power a single nerve fibre: note

- The medullary sheath: stained black in consequence of its fatty nature: interrupted at intervals by
- ii. The nodes of Ranvier: spots where the medullary sheath is absent, and the primitive sheath forms constrictions touching

- iii. The axis cylinder: a central cylindrical rod, the essential part of the nerve fibre: visible at the nodes: much less deeply stained than the medullary sheath.
- iv. Nuclei: project into the medullary sheath about midway between the nodes.

Tease a small piece of fresh nerve in chloroform: this will partially dissolve the fatty medullary sheath and so render clearly visible

- i. The primitive sheath, or sheath of Schwann.
- ii. The axis cylinder.

#### b. Non-medullated nerve fibres.

Examine mounted preparation of frog's cornea stained with chloride of gold: note the fine varicose nerve fibres with no medullary sheath.

- 2. Nerve cells: Tease in glycerine a small fragment of the anterior cornu of the spinal cord of the ox (lumbar region): cover, and examine with low and high powers: note
  - The nerve cells: large nucleated cells with many branching arms.
  - ii. The nerve fibres.
  - The fine connective tissue binding the several parts together.

#### 3. Structure of the Spinal Cord.

Take one of the pieces of spinal cord of frog stained and imbedded in spermaceti: cut transverse sections of it in the manner described in the Introduction, keeping oil on the razor and clearing the sections with creosote and turpentine. When quite clean, mount in balsam, and examine with low and high powers: note

## a. With low power.

- Shape: bilaterally symmetrical; considerably wider than it is deep.
- ii. The anterior and posterior fissures.

- iii. The white matter: forming the outer part of the corf, and composed of medulated nerve flores.
- iv. The grey matter: forming the central part of the cent, and composed of a dense network of non-medulated nerve filtres, in which are imbedded numerous nerve cells.
- v. The cornect: anterior or ventral, and posterior or dorsal, into which the every matter is produced on either side.
- vi. The central canal of the cord: in the median line, nearer the ventral than the dural surface.
- vii. The nerve rusts: only seen if the section happens to pass through their more of origin.
  - a. The posterior or derail root: connected with the posterior esent of the grey matter: a single thick hand of perve fibres.
  - pl. The anterior or ventral rost: connected with the anterior cornu of the grey matter: a number of very slender bands of nerve firms.

#### h. With high power.

- i. The ganglion cells: large uncleated branched cells lying in groups in the grey matter: largest and most unmerous in the anterior comma.
- ii. The neuroglia or delicate network of fibres and cells penetrating and supporting all parts of the cord, and continuous at the surface with
- iii. The pia mater, the delicate connective tissue membrane ensheathing the cord and extending into the fusures.
- iv. The bloodvessels of the cord: small and numerous: enter from the pia mater.
- v. The ciliated epithelium lining the central canal.

#### Chap. VII.—THE EYE.

## A. The Eye of the Frog.

Remove the eye from a freshly killed frog: snip off with scissors the muscles of the eyeball: note

- The shape of the eyeball: flattened on the outer side: more convex on the inner or deeper side.
- ii. The sclerotic: the firm outer wall of the eyeball, formed of dense white connective tissue strengthened by hyaline cartilage.
- iii. The cornea: the transparent patch on the outer side of the eye through which the light enters: continuous at its margin with the sclerotic.
- iv. The iris: a pigmented ring placed behind the cornea and seen through it: acts as a diaphragm, limiting the amount of light that enters the eye.
- v. The pupil: the elliptical aperture surrounded by the iris, and serving to admit the light to the interior of the eye.
- vi. The optic nerve: seen piercing the sclerotic to enter the eyeball on its inner side.

Place the eye under water and divide it with scissors into two halves by a cut passing through the middle of the cornea and through the sclerotic close to the optic nerve, so as to lay open completely the interior of the eye: note

- The lens: a firm solid transparent body, just behind the iris and attached to its outer margin. More convex on its posterior than its anterior surface.
- ii. The anterior chamber of the eye: between the lens and cornea: small: contains the aqueous humour.
- iii. The posterior chamber of the eye: the large space behind the lens: filled by the vitreous humour, a gelatinous body.

- iv. The choroid: the black pigmented layer lining the sclerotic, and continuous in front with the iris.
- v. The retina: a delicate transparent membrane lining the posterior two-thirds of the eye: is readily detached from the choroid, except at the entrance of the optic nerve with which it is continuous.

#### B. The Eye of the Sheep or Ox.

Dissect off the muscles of the eyeball and the fat which surrounds the optic nerve: note

- i. The shape of the eyeball.
- The sclerotic, covering about five-sixths of the eyeball; tough, white, and opaque.
- iii, The cornea: covering the anterior sixth of the eyeball: circular, transparent: continuous at its margin with the sclerotic: more convex than the sclerotic.
- iv. The conjunctiva: a delicate epithelial layer, covering the front of the cornea and part of the sclerotic.
- v. The iris: a pigmented ring seen through the cornea.
- vi. The pupil: the central aperture surrounded by the iris.
- vii. The optic nerve: piercing the sclerotic at the back of the eye.

Cut all round the cornea close to its margin with fine scissors: remove the cornea: note

- The aqueous humour: the transparent watery fluid filling the anterior chamber of the eye, and escaping when the cornea is removed.
- ii. The lens.

Make four radial cuts equidistant from one another through the margin of the cornea and the sclerotic, taking care not to injure the deeper parts; and extend the cuts back towards the optic nerve. Carefully peel off the four flaps into which the sclerotic is now divided from the underlying black choroid coat: turn them down, and pin them to the dissecting board so as to fix the eye with the iris upwards: note

- i. The ciliary muscle: a whitish ring of unstriped muscle connecting the outer margin of the iris with the junction of the cornea and sclerotic: this must be detached with the handle of the scalpel to allow the flaps of the sclerotic to be turned back.
- The choroid: the dense black coat exposed by the removal of the sclerotic.
- The ciliary vessels: pierce the sclerotic to convey blood to and from the very vascular choroid.
- iv. The ciliary nerves: seen passing through the sclerotic to the choroid while the flaps are being turned down.

Make a couple of radial incisions a short distance apart through the iris and ciliary muscle, and turn back the portion of the iris between the two cuts: note

> i. The ciliary processes: a series of radial folds on the under surface of the outer margin of the iris: they fit into corresponding folds in the ligament which surrounds and supports the lens.

Make a circular incision with scissors all round the eye about a quarter of an inch behind the ciliary muscle through the choroid and retina, but taking care not to injure the delicate membrane enclosing the vitreous humour. Gently detach and remove in one piece this front ring of iris, choroid, and retina: pin it on the dissecting board with the posterior surface upwards: note

- i. The ciliary processes.
- ii. The uvea: the layer of dense black pigment at the back of the iris.
- iii. The ora serrata: the indented anterior border of the retina, which extends forwards nearly as far as the ciliary processes.

iv. The ligamentum pectinatum iridis: a series of small festoon-like processes, connecting the posterior surface of the cornea with the outer margin of the iris: to see these the ring must be turned over and the iris examined from in front.

Examine the front of the eyeball from which the iris has been removed: note

- i. The capsule of the lens: transparent and elastic.
- ii. The suspensory ligament of the lens, or zonule of Zinn: the outer margin of the capsule of the lens: marked with radiating folds into which the ciliary processes fit.
- iii. The hyaloid membrane: the delicate elastic membrane enclosing the vitreous humour, and continuous in front with the posterior layer of the capsule of the lens.
- iv. The cut edges of the retina and choroid.

Remove the lens from its capsule: note its shape, more convex behind than in front: harden it by spirit, or by boiling for a few minutes.

Remove the vitreous humour in the hyaloid membrane: note

- i. The retina: a delicate pulpy membrane between the hyaloid membrane and the choroid.
- ii. The blind spot: the point of entrance of the optic nerve: to this spot the retina adheres firmly, though it can be readily removed from the choroid at all other parts.
- iii. The retinal vessels: radiating from the blind spot.

# C. Histology of the Eye.

#### 1. The frog's eye.

Mount in balsam one of the prepared sections of the posterior part of the frog's eye: examine with low and high powers: note

- a. The sclerotic: consisting chiefly of hyaline cartilage.
- b. The choroid: a vascular plexus with much pigment.

- c. The retina: composed of the following layers from without inwards:
  - i. Layer of pigment cells, sending processes between
  - ii. The rods and cones: a single layer of columnar bodies placed vertically to the surface: the rods are far more numerous, and much larger than the cones: each consists of an inner and an outer segment.
  - The outer nuclear layer: a moderately thick layer, well stained.
  - iv. The outer molecular layer: a very thin layer, not stained.
  - v. The inner nuclear layer; thick, and well stained.
  - vi. The inner molecular layer: thick, but not stained.
  - vii. The layer of nucleated nerve cells.
  - viii. The layer of nerve fibres.
  - ix. The internal limiting membrane.
  - x. The radial fibres: commencing with expanded ends in the ninth layer and stretching outwards: can easily be traced through the inner molecular layer.
- 2. The choroid: spread a small piece of fresh choroid on a slide in normal salt solution: examine with low and high powers: note
  - i. The network of bloodvessels: invested by
  - ii. Pigment cells: irregularly branched: with clear nuclei.
- 3. The lens: tease in glycerine a small piece of lens hardened by boiling: examine with low and high powers: note
  - i. The elongated cells of which the lens is composed.
  - ii. The serrated edges of many of the cells.

# Chap. VIII.—THE REPRODUCTIVE ORGANS AND THE CLOACA OF THE FROG.

#### A. The Male Frog.

#### 1. The Reproductive organs.

- i. The testes: a pair of yellow oval bodies about a quarter of an inch long, lying on the ventral surface of the kidneys. Form the essential male elements or spermatozoa.
- ii. The vasa efferentia: a number—usually 10 to 12—of slender ducts, connecting the testis of each side with the inner or median border of the corresponding kidney: serve to convey the spermatozoa from the testis into the tubules of the kidney, from which they escape by the ureter which acts as
- iii. The vas deferens: runs along the outer side of the posterior part of the kidney and then back to the cloaca.
- iv. The vesicula seminalis: a large pouch-like dilatation on the outer side of the vas deferens, just below the kidney and before reaching the cloaca.

#### 2. The Cloaca.

Lay the frog on its back: cut through the femur on each side with strong scissors just below its head: cut also through the two ilia at their necks just in front of the acetabula. Carefully dissect away the pelvic symphysis, which is now isolated, from the muscles attached to it and from the cloaca, and remove it completely, taking care not to injure the cloaca.

- The cloaca: really the terminal portion of the large intestine into which open the ureters, the genital ducts, and the bladder.
- ii. The bladder: a thin-walled bifid muscular sac, lying on the ventral side of the large intestine and cloaca, the two halves communicating

freely together. It is invested by peritoneum and attached to the sides of the body by special peritoneal folds.

Inflate the bladder through the cloacal aperture: pass a seeker up the cloaca to determine the exact position of the opening from the bladder to the cloaca. Cut up the cloaca along one side: wash out its contents and examine the opening into the bladder.

iii. The ureter or vas deferens below the vesicula seminalis forms a very short tube opening into the dorsal wall of the cloaca almost exactly opposite the opening of the bladder on the ventral surface. The openings of the two ureters are close together on the apices of two small papillæ, overhung by a slight valvular projection of the mucous membrane of the cloaca.

#### B. The Female Frog.

#### 1. The Reproductive organs.

- i. The ovaries: a pair of black masses lying in folds of peritoneum in front of the kidneys, in very much the same position as the testes in the male. Their shape and size vary much at different seasons of the year. On their surfaces are numerous rounded projections, like small shot; these are ova in various stages of development: the smaller and younger ones are white; the larger and more mature ones black in one half, and white or yellowish in the other.
- ii. The oviducts: a pair of white much convoluted tubes with thick gelatinous walls. Commence with open mouths at the extreme front end of the body cavity, close to the outer side of the roots of the lungs; and run back increasing in size, and becoming much convoluted. The lower ends are much dilated, but have thinner walls. Unlike the male the genital ducts are distinct from the ureters.

#### 2. The cloaca.

Dissect as in male.

- i. The cloaca: very similar to that of the male.
- ii. The bladder: like that of the male.
- iii. The **oviducts:** open separately into the dorsal wall of the cloaca just above the bladder by two wide apertures separated by a narrow median partition.
- iv. The ureters: open by two small apertures placed close together into the dorsal wall of the cloaca just below the oviducts.

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